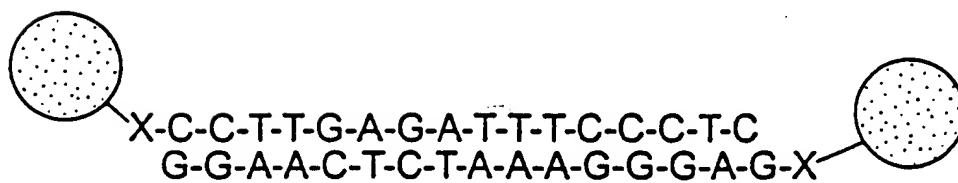
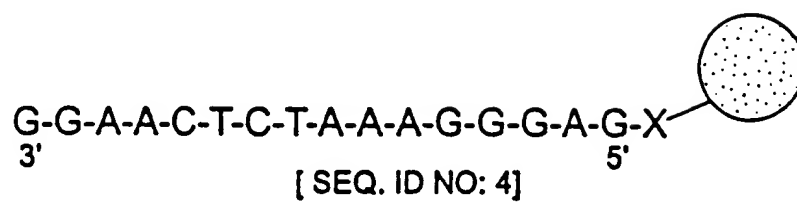
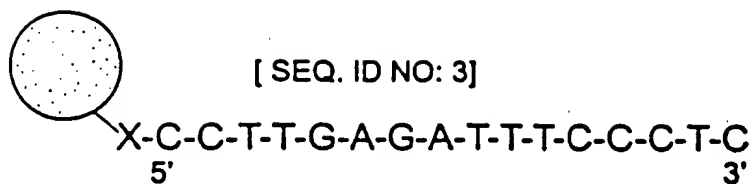


FIG. 1



TTTTT-4854660

FIG. 2

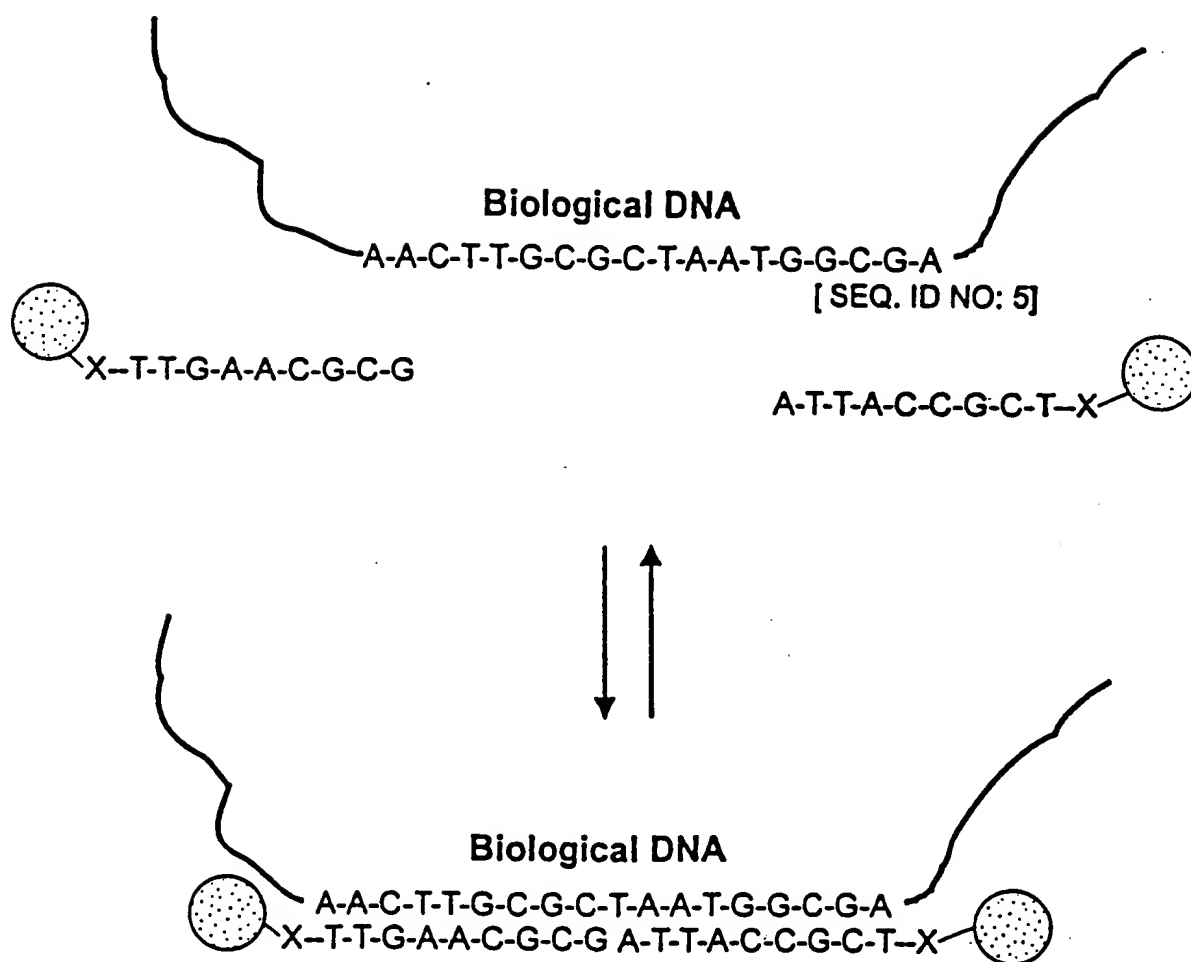


FIG.3

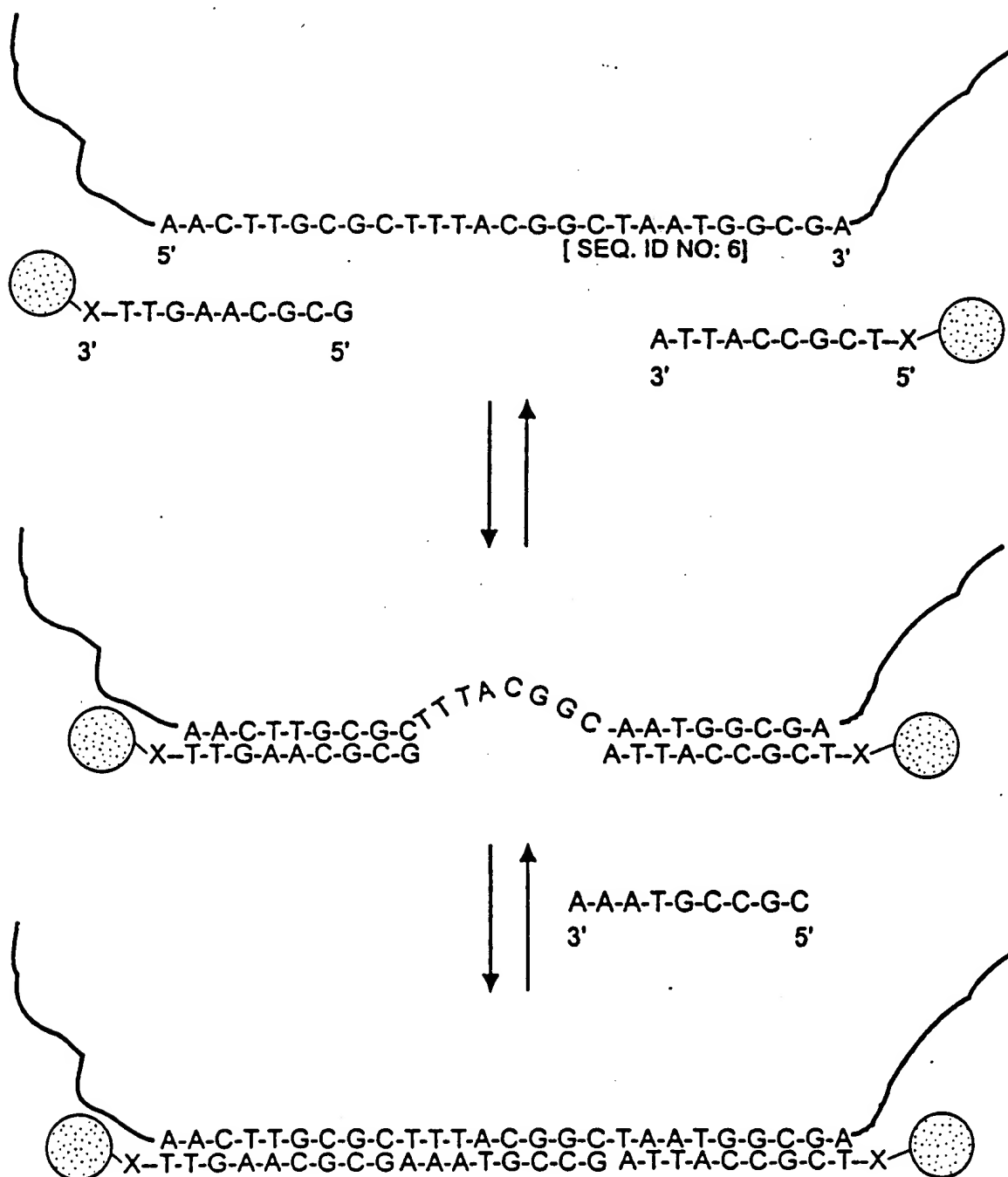
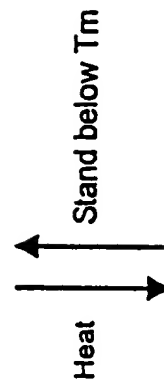
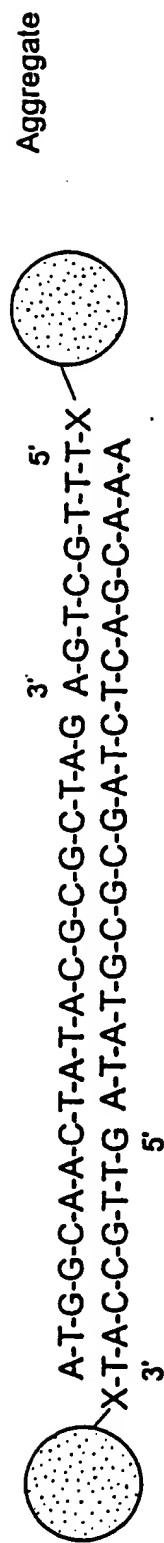
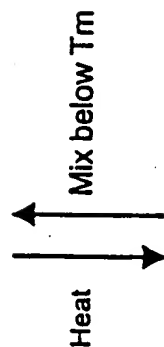
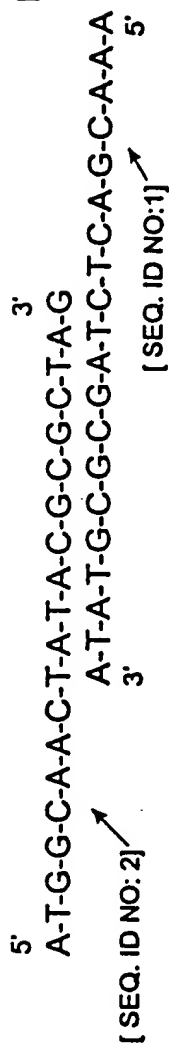


FIG. 3

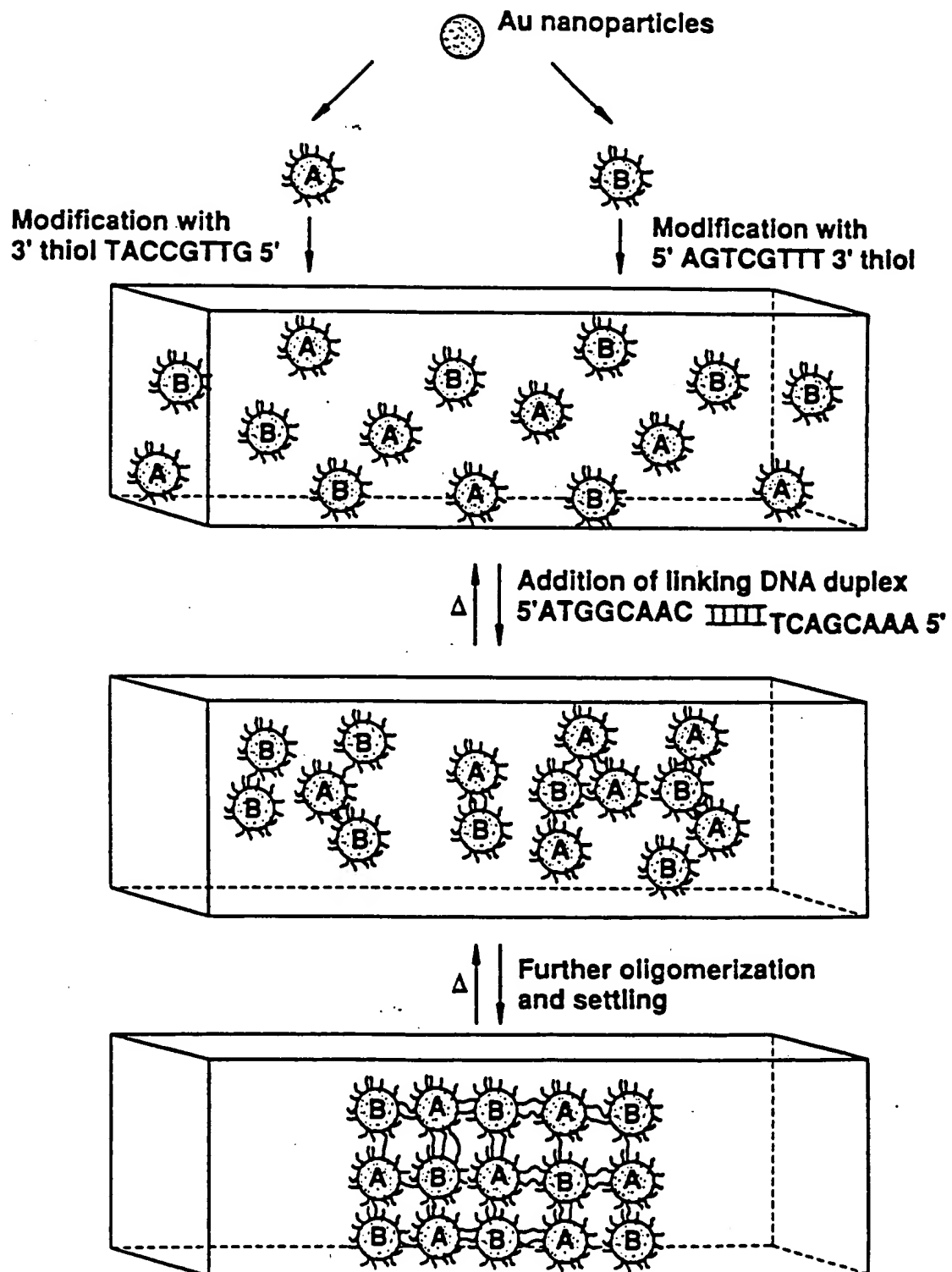
FIG. 4

Linking oligonucleotide



Precipitate (formed by further cross-linking)

FIG.5



TOP SECRET



FIG. 6A



FIG. 6B



FIG. 6C

FIG. 7

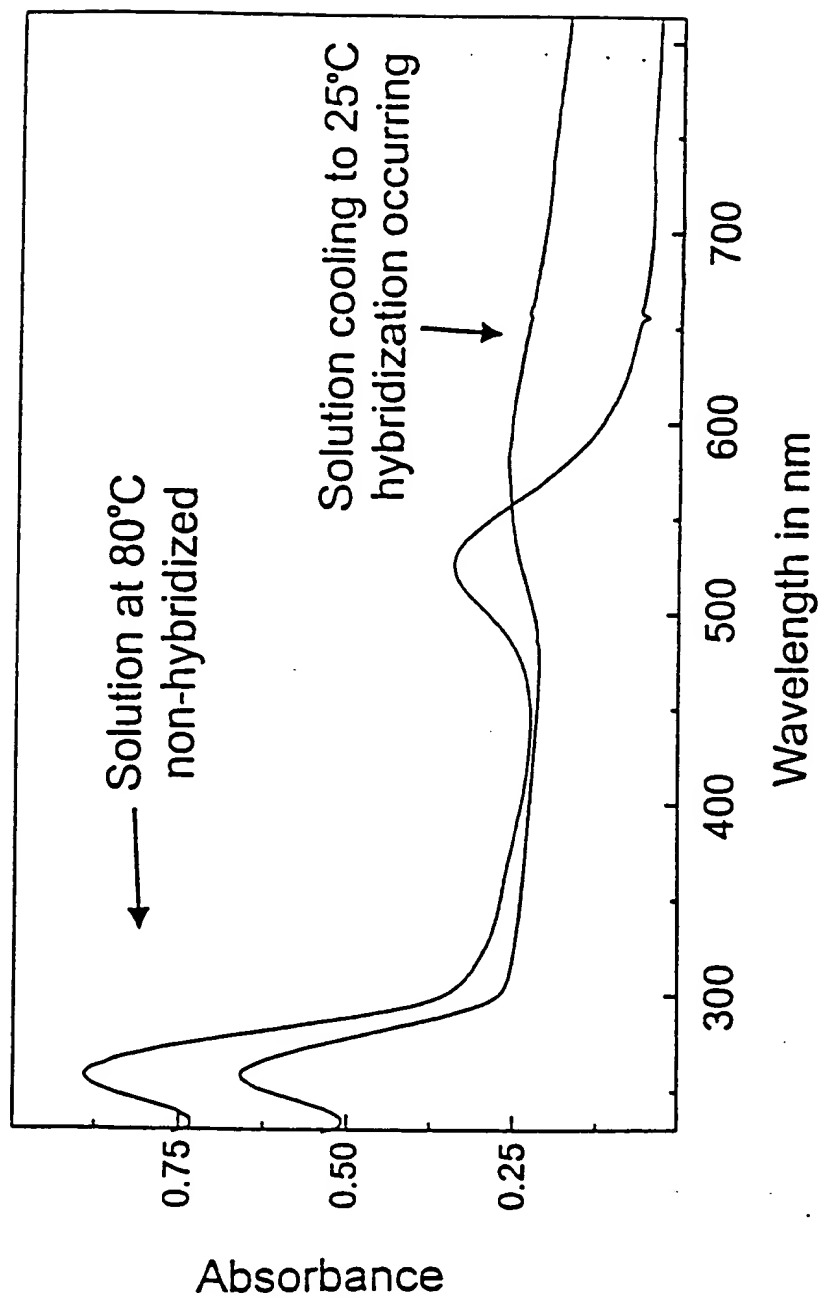


FIG.8A

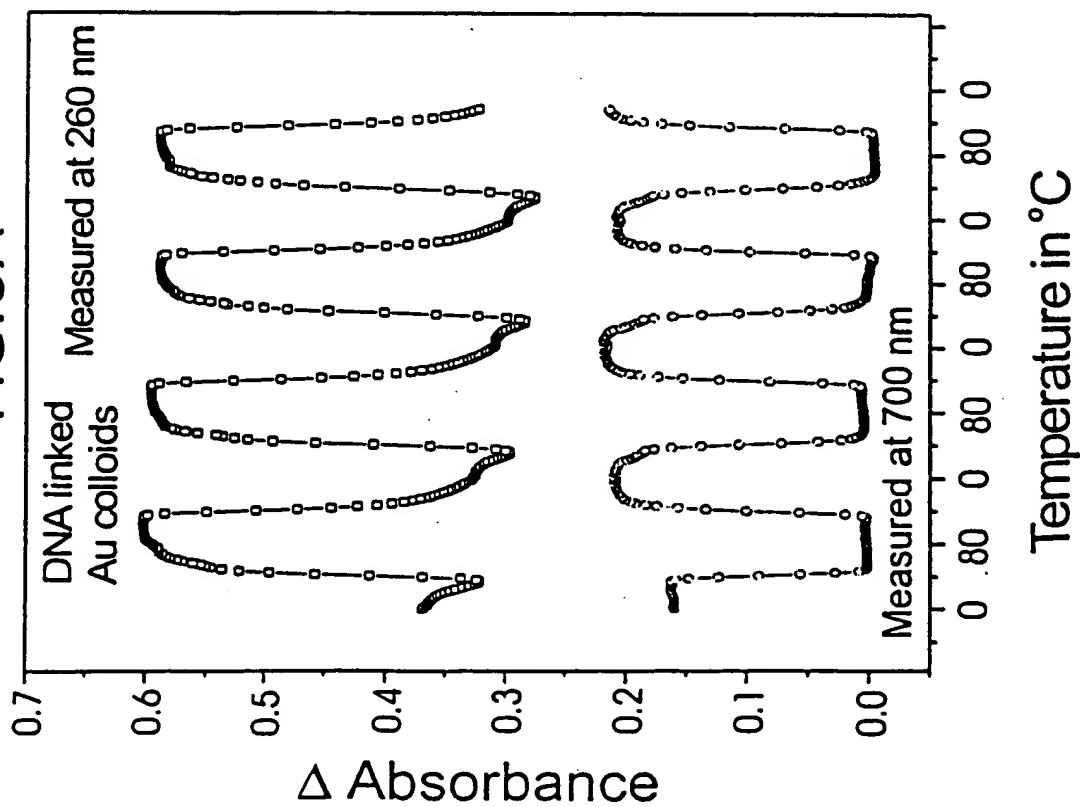
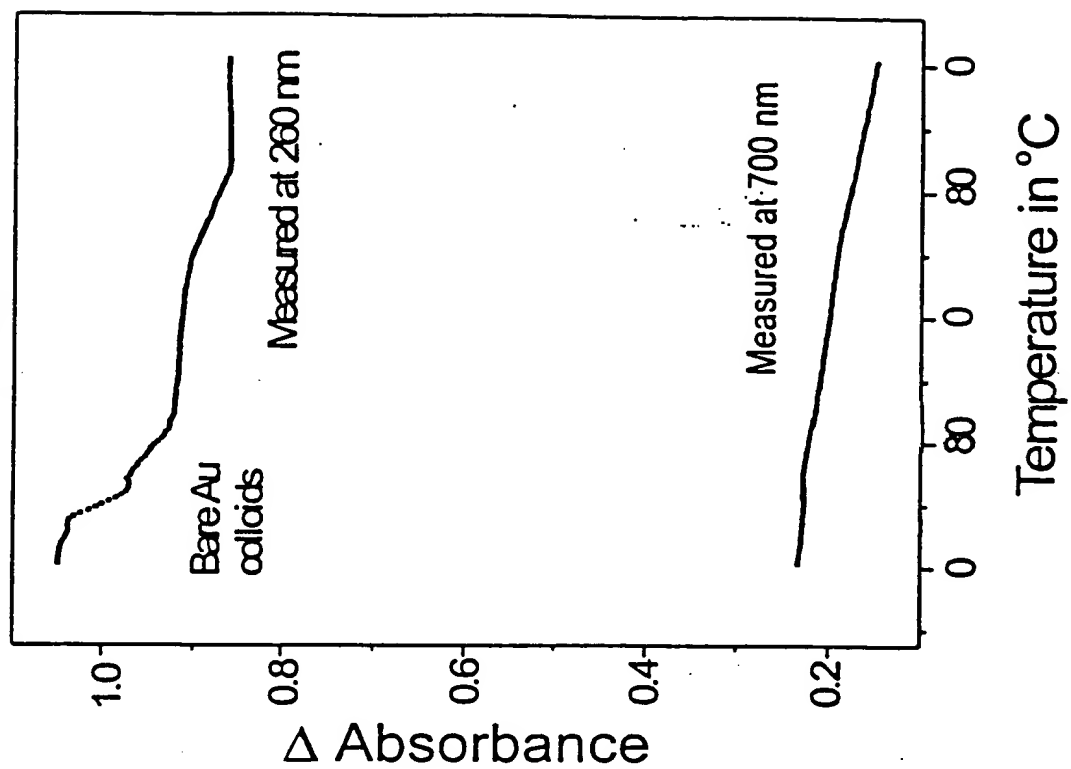


FIG.8B



099534-4BES/660



FIG.9A

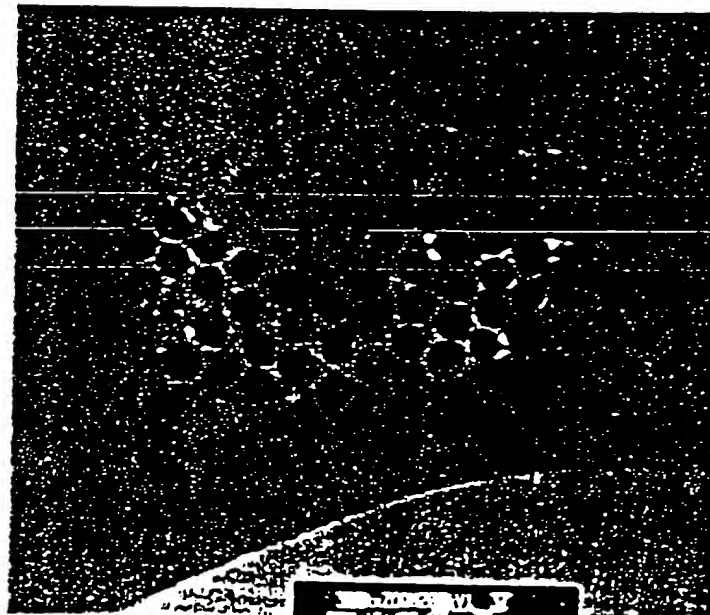


FIG.9B

FIG.10

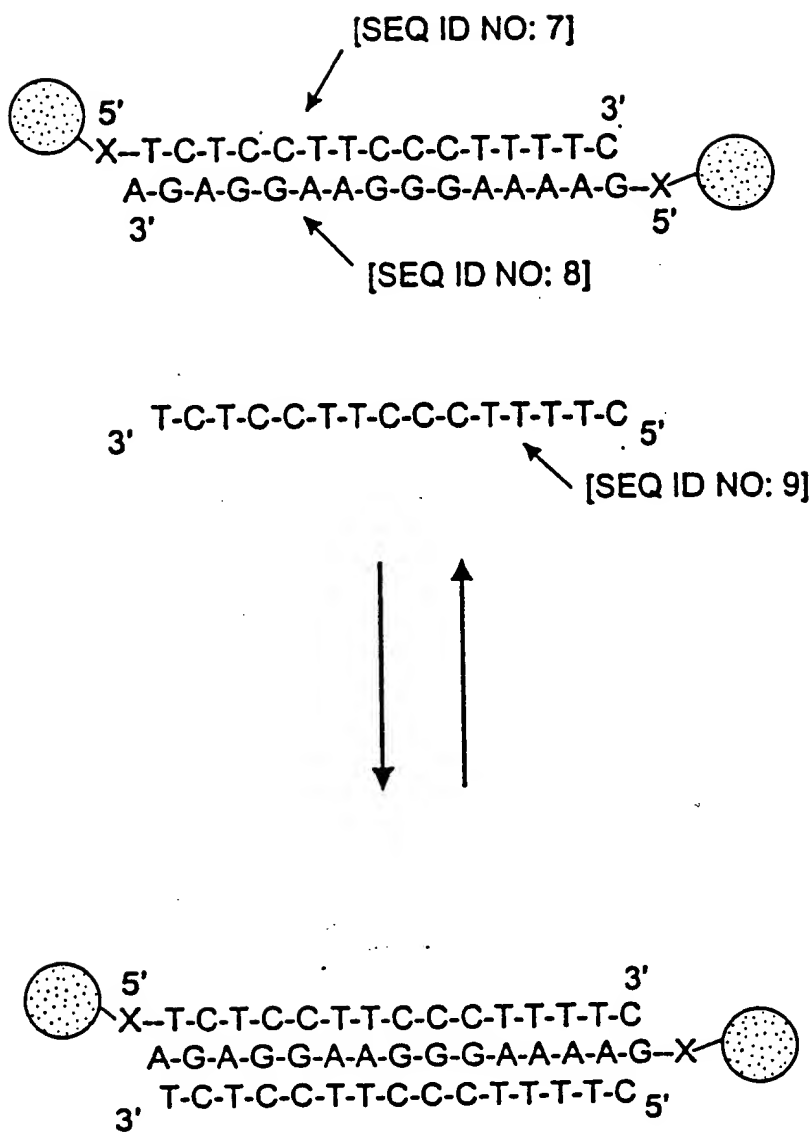


FIG.11

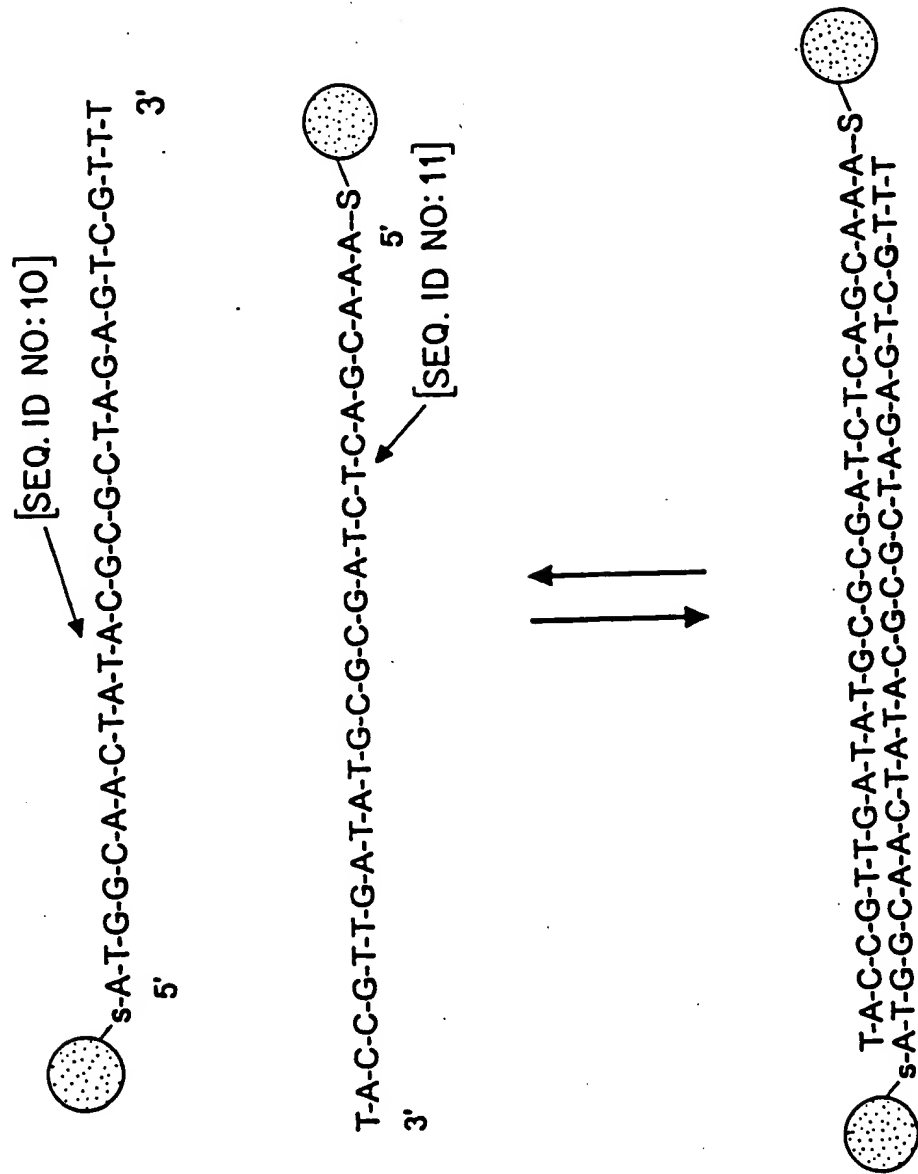


FIG. 12A

Complementary Target

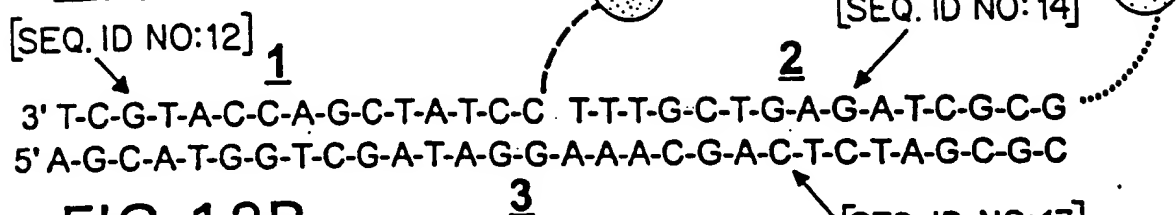


FIG. 12B

Probes without Target



FIG. 12C

Half Complementary Target

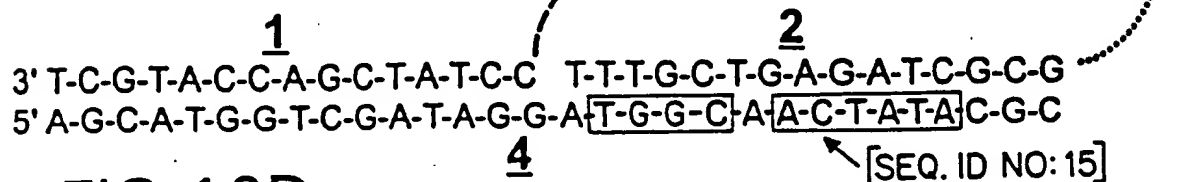


FIG. 12D

Target - 6 bp

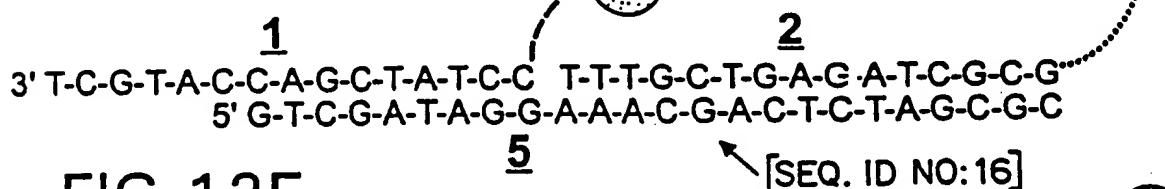


FIG. 12E

One bp Mismatch

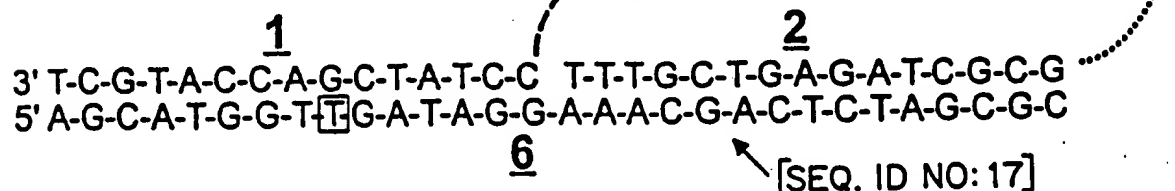
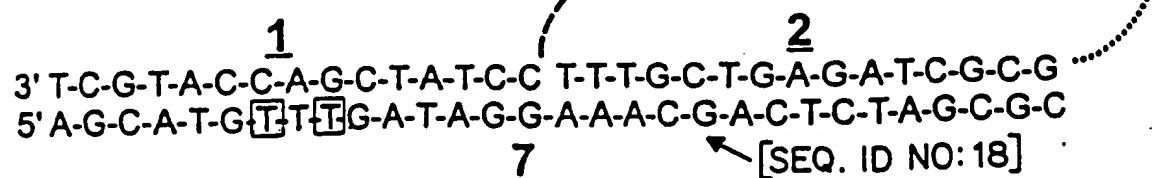


FIG. 12F

Two bp Mismatch



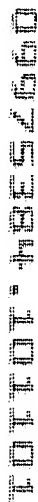


FIG.13B

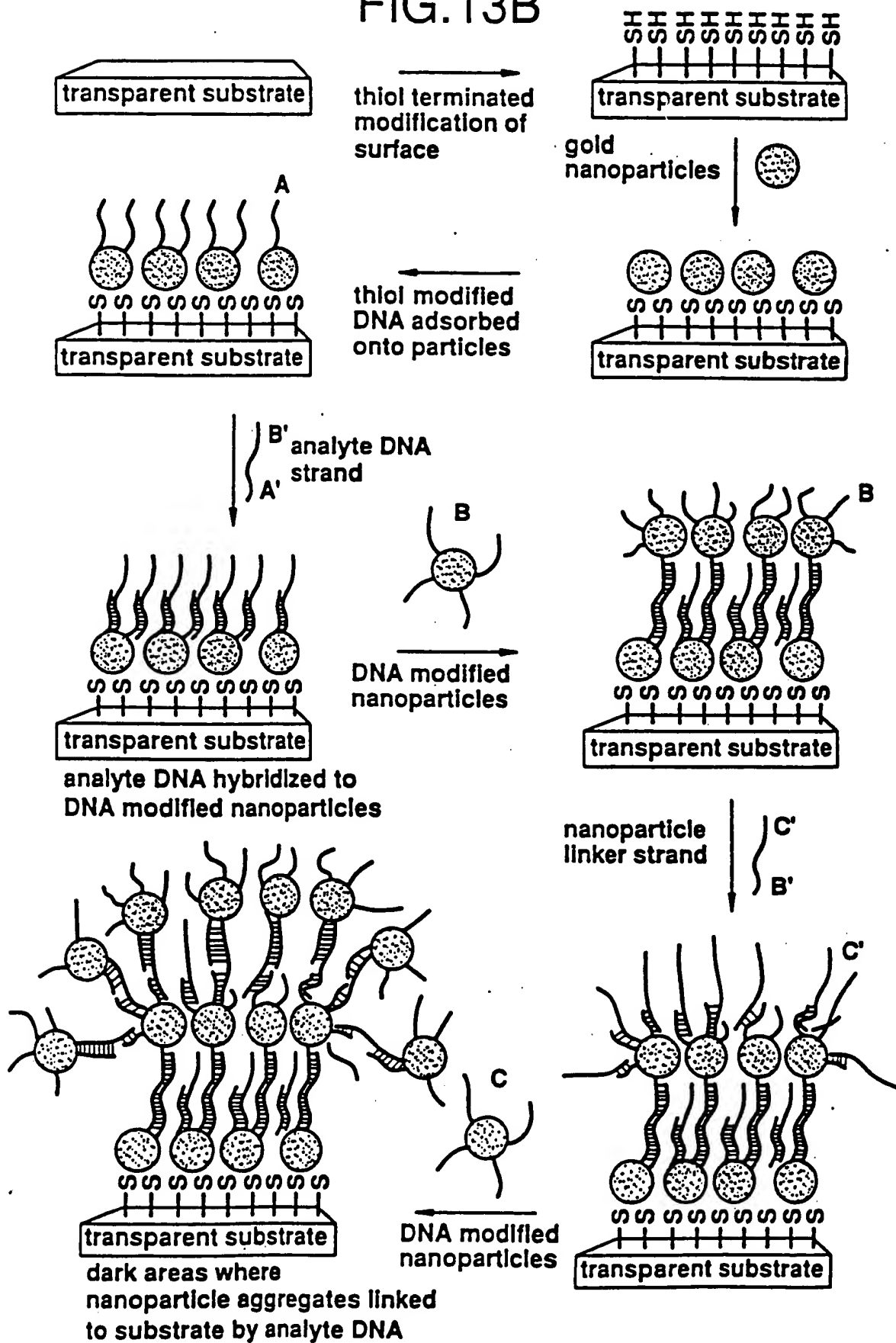


FIG.14A

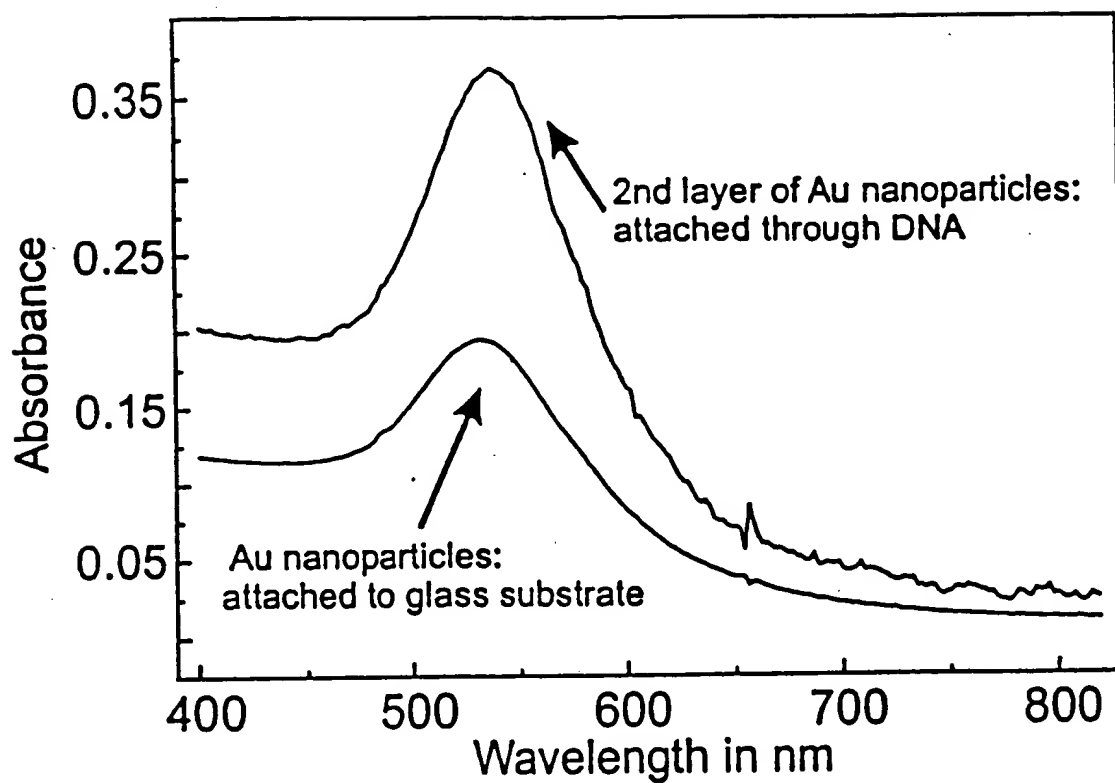
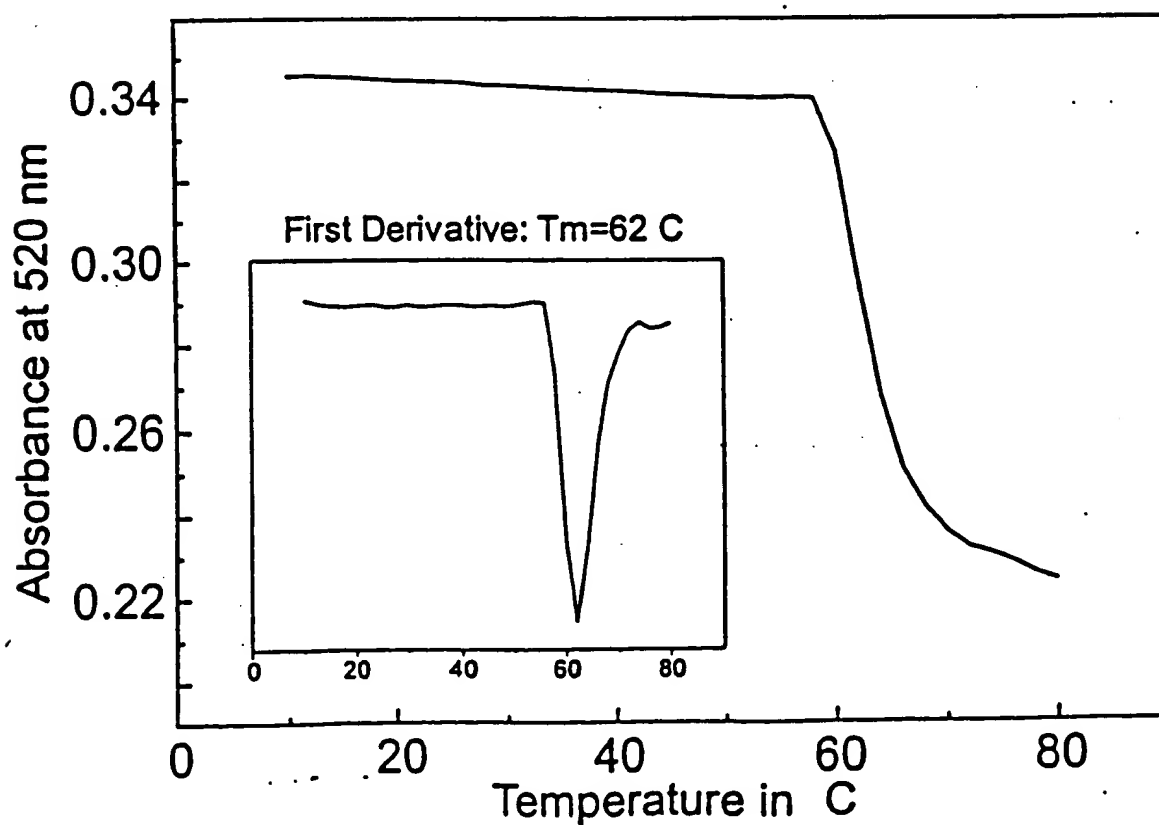


FIG.14B



Probes with No Target

FIG15B

Half-Complementary Target

FIG15C

Complementary Target

T_m=53.5°C

FIG15D

ONE Base-Pair Mismatch at Probe Head

T_m=50.4°C

FIG15E

ONE Base-Pair Mismatch at Probe Tail

T_m=46.2°C

FIG15F

ONE Base Deletion

T_m=51.6°C

FIG15G

ONE Base-Pair Insertion

T_m=50.2°C

SEQ ID NO:26

5' TAC-GAG-TTG-AGA-CAT-CCT-GAA-TGC-G 3'

S-ATG-CTC-AAC-TCT TA-GGA-CTT-ACG-C-S

1 2

FIG. 16A

24 Base Template

5' TAC-GAG-TTG-AGA-ATC-CTG-AAT-GCG 3'
—S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S —

—S-ATG-CTC-AAC-TCT TAG-GAC-TTA-CGC-S —

१

2

FIG. 16B
48 Base Template with Complementary 24 Base Filler

5' TAC-GAG-TTG-AGA-CCG-TTA-AGA-CGA-GGC-AAT-CAT-GCA-ATC-CTG-AAT-GCG 3' /
-S-ATG-CTC-AAC-TCT GGC-AAT-TCT-GCT-CCG-TTA-GTA-CGT TAG-GAC-TTA-CGC-S-

—S-ATG-CTC-AAC-TCT GGC-AAT-TCT-GCT-CCG-TTA-GTA-CGT TAG-GAC-TTA-CGC-S —

1

21

FIG. 16C
72 Base Template with Complementary 48 Base Filler

5' TAC-GAG-TTG-AGA-CCG-TTA-AGA-CGA-GGC-AAT-CAT-GCA-TAT-ATT-GGA-CGC-TTT-ACG-GAC-AAC-ATC-CTG-AAT-GCG 3' —S-ATG-CTC-AAC-TCT GGC-AAT-TCT-GCT-CCG-TTA-GTA-CGT-ATA-TAA-CCT-GCG-AAA-TGC-CTG-TTG TAG-GAC-TTA-CGC-S—

—S-ATG-CTC-AAC-TCT GGC-AAT-TCT-GCT-CCG-TTA-GTA-CGT-ATA-TAA-CCT-GCG-AAA-TGC-CTG-TTG TAG-GAC-TTA-CGC-S—

१।

21

FIG.17A

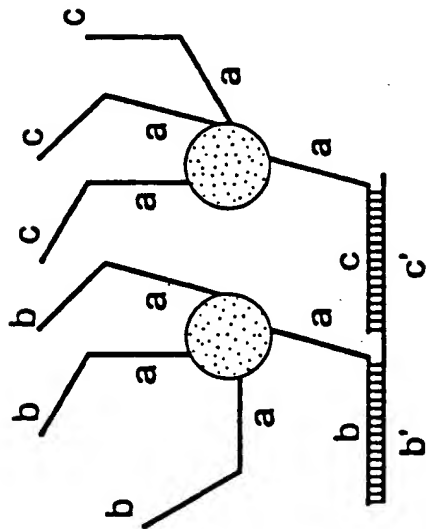


FIG.17B

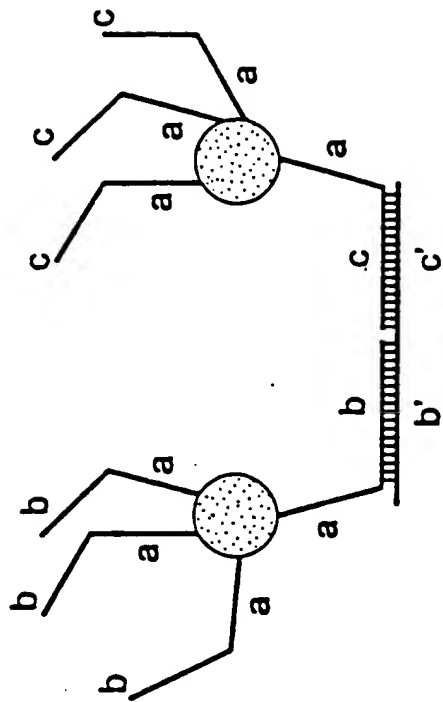


FIG.17C

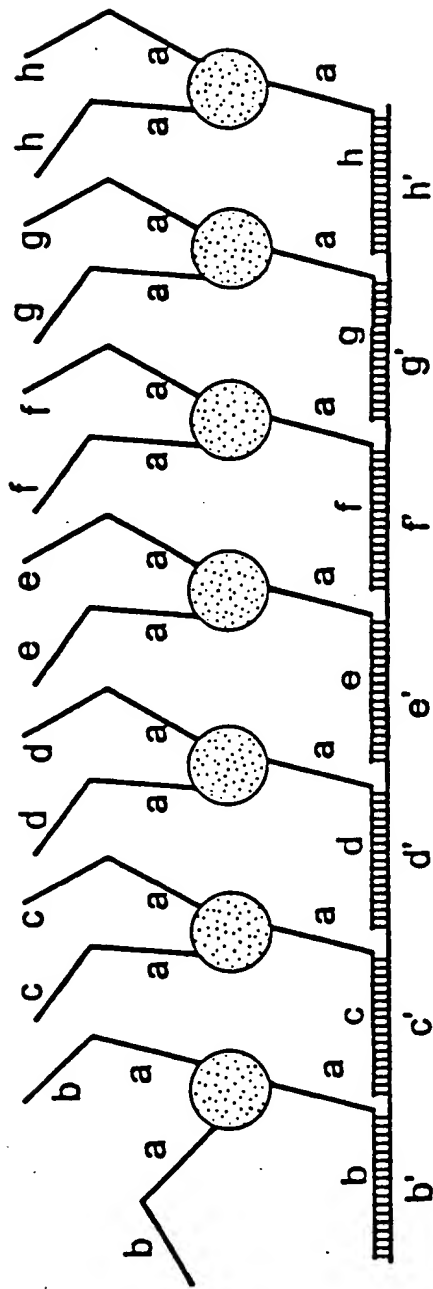


FIG.17D

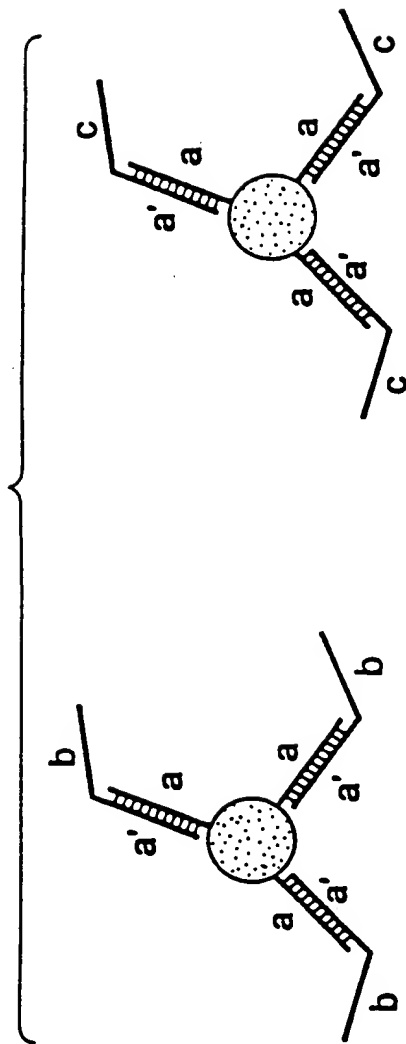


FIG.17E

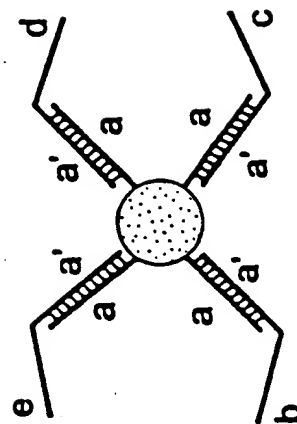


FIG.18

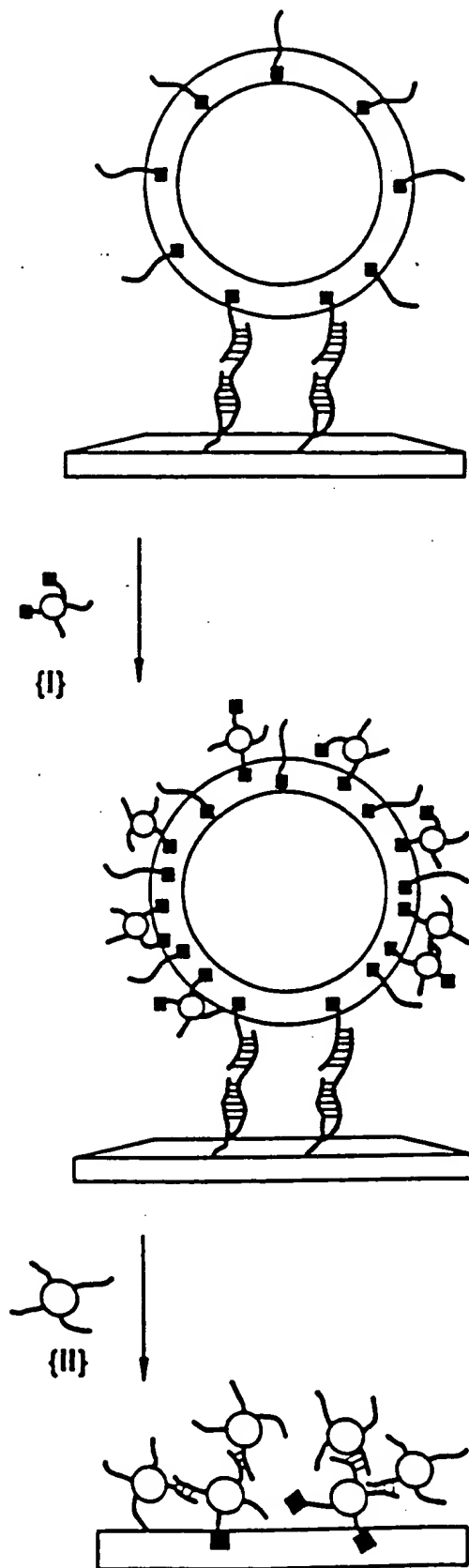


FIG.19A

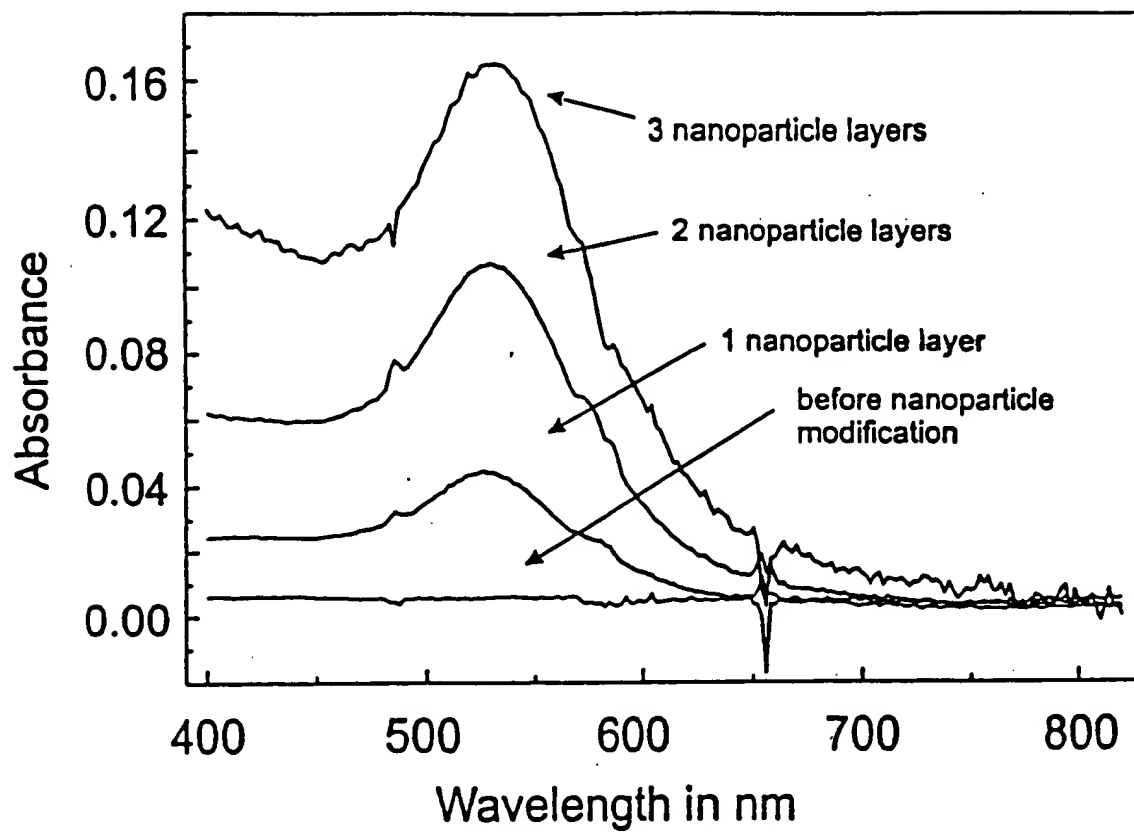
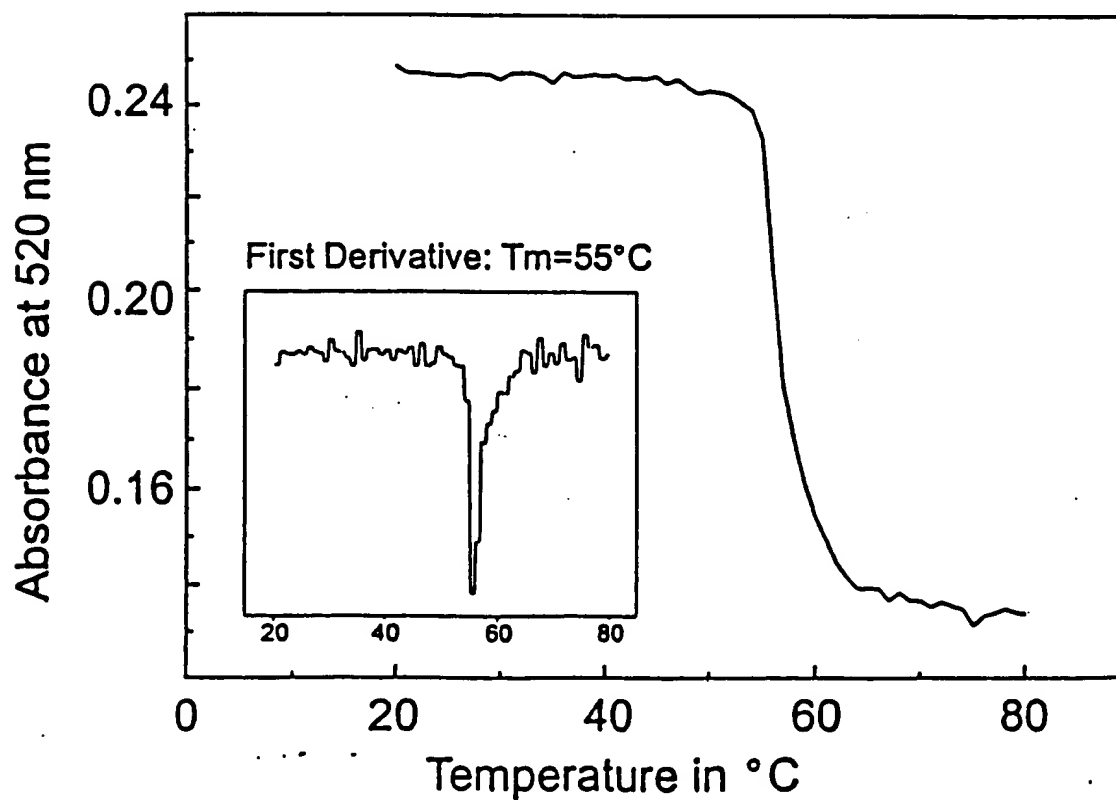


FIG.19B



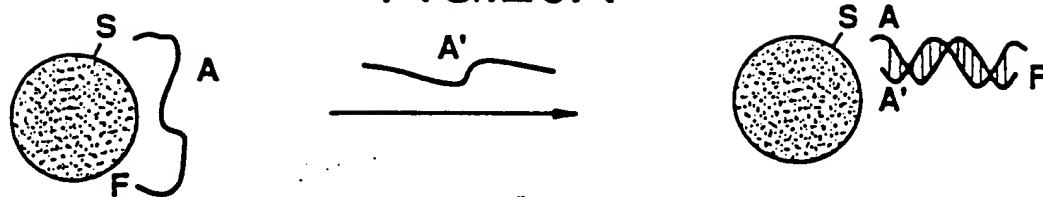
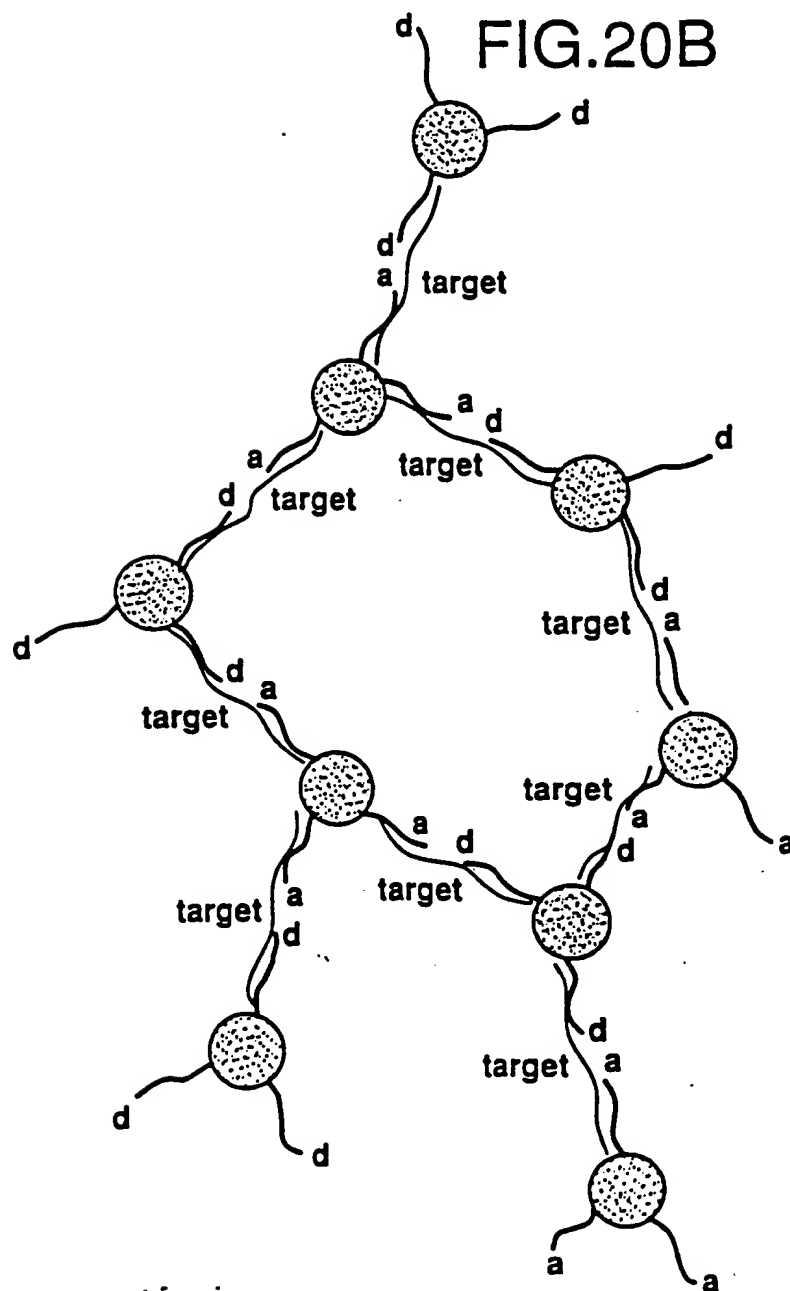
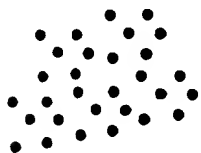


FIG.20B



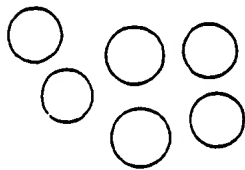


Oligonucleotide
modified Au
nanoparticle probes



Red

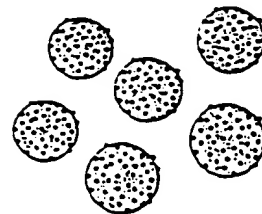
Fluorophore labeled
oligonucleotide modified
latex probes



White/Fluorescent

Target
Oligonucleotide

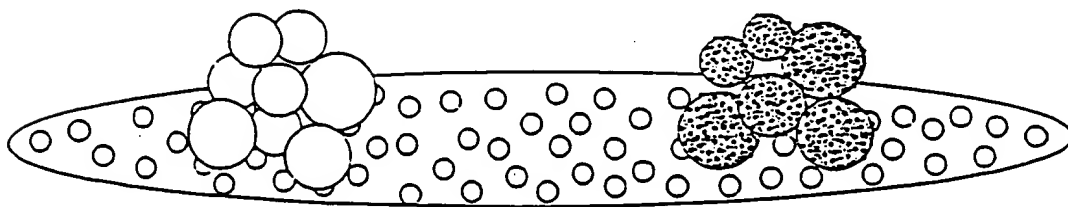
Au/Latex hybrid



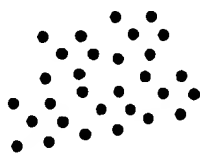
Pink/Non-fluorescent

No Target
Oligonucleotide

Target
Oligonucleotide



All Au probes pass
through membrane

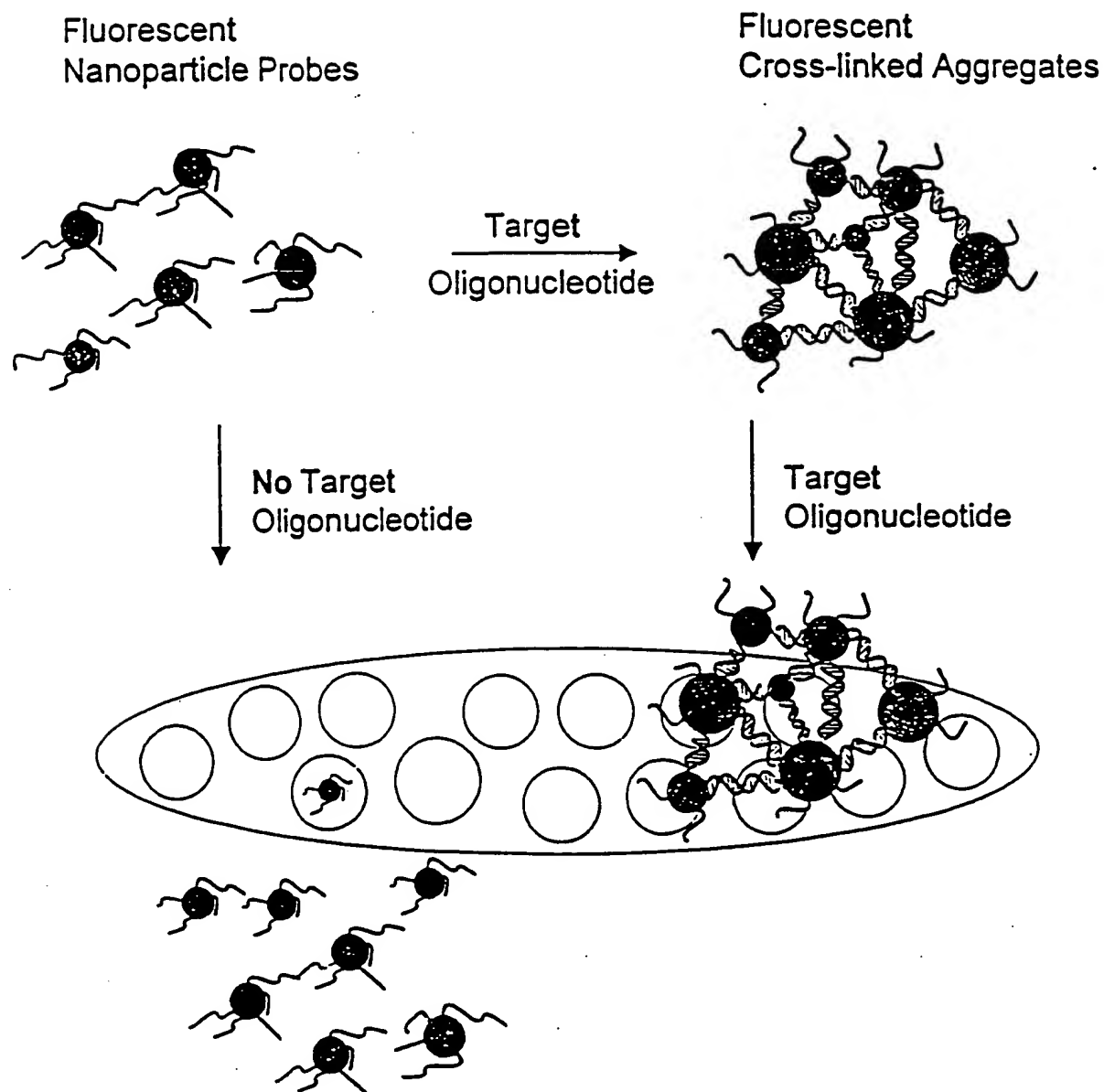


Excess Au probes
pass through
membrane



FIGURE 21

FIGURE 22



The fluorescent nanoparticle probes pass through the membrane

The fluorescent cross-linked aggregates are retained by the membrane

Anthrax PCR Product

5'G GCG GAT GAG TCA GTA GTT AAG GAG GCT CAT AGA GAA GTA ATT AAT
3'C CGC CTA CTC AGT CAT CAA TTC CTC CGA GTA TCT CTT CAT TAA TTA

TCG TCA ACA GAG GGA TTA TTG TTA AAT ATT GAT AAG GAT ATA AGA AAA
AGC AGT TGT CTC CCT AAT AAC AAT TTA TAA CTA TTC CTA TAT TCT TTT

ATA TTA TCC AGG GTT ATA TTG TAG AAA TTG AAG ATA CTG AAG GGC TT 3'
TAT AAT AGG TCC CAA TAT AAC ATC TTT AAC TTC TAT GAC TTC CCG AA 5'

141 mer Anthrax PCR product [SEQ ID NO:36]

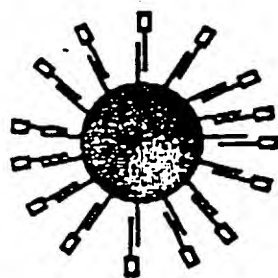
3' CTC CCT AAT AAC AAT [SEQ ID NO:37]
3' TTA TAA CTA TTC CTA [SEQ ID NO:38]
Oligonucleotide-Nanoparticle Probes

Blocker Oligonucleotides

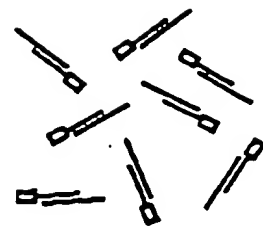
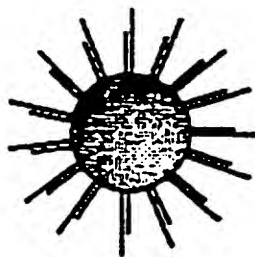
3' C CGC CTA CTC AGT CAT CAA TTC CTC CGA GT [SEQ ID NO:39]
3' A TCT CTT CAT TAA TTA AGC AGT TGT [SEQ ID NO:40]
3' TAT TCT TTT TAT AAT AGG TCC CAA TAT [SEQ ID NO:41]
3' AAC ATC TTT AAC TTC TAT GAC TTC CCG AA [SEQ ID NO:42]

FIGURE 23

TOTAL: 4352660



Satellite Probe



Detection Signal

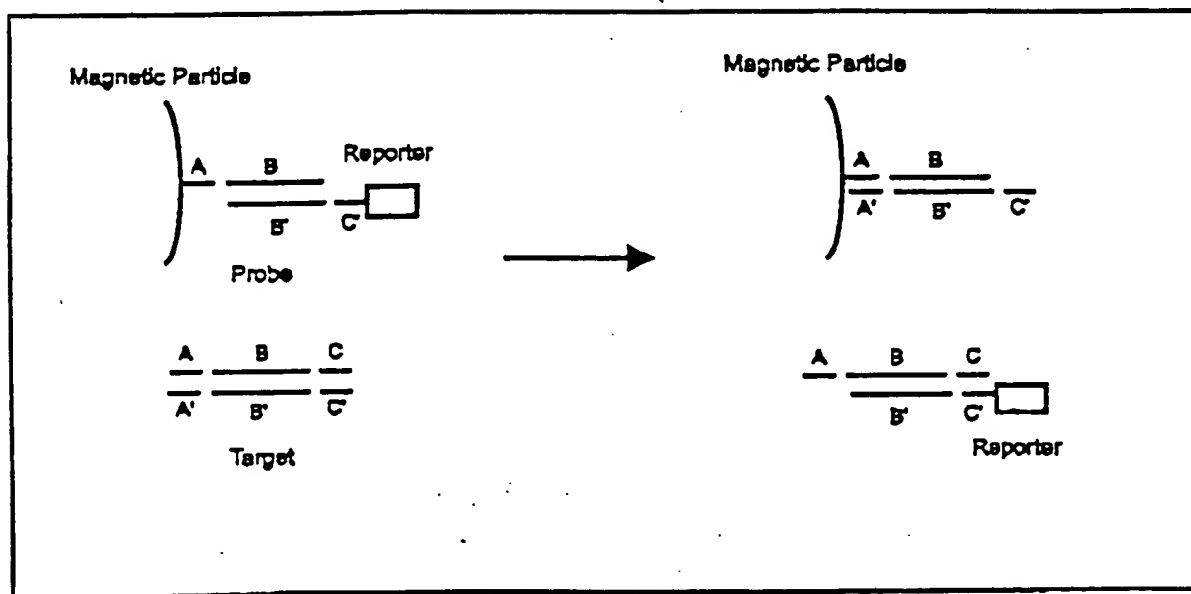


FIGURE 24

70707-4352660

TTTTT" +BESZ660

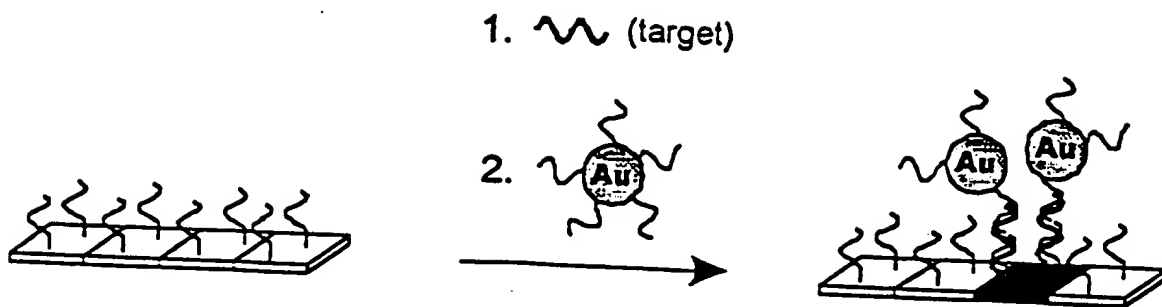


FIGURE 25A

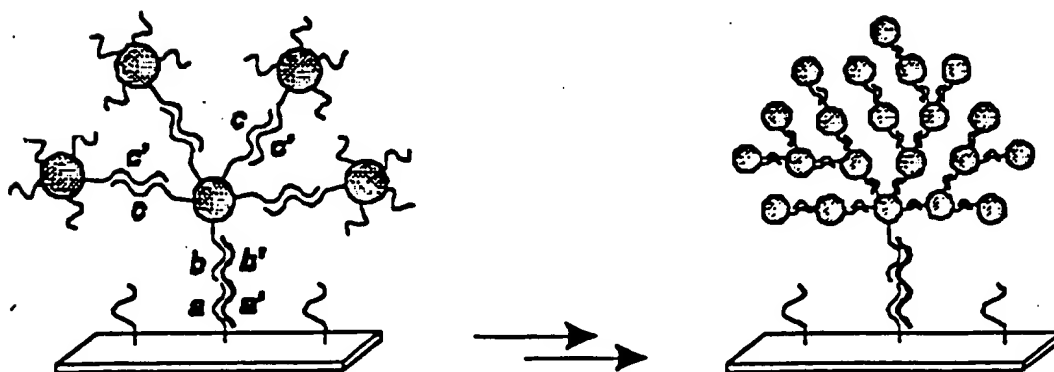
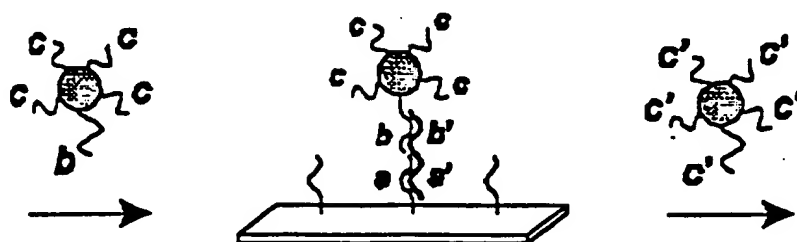
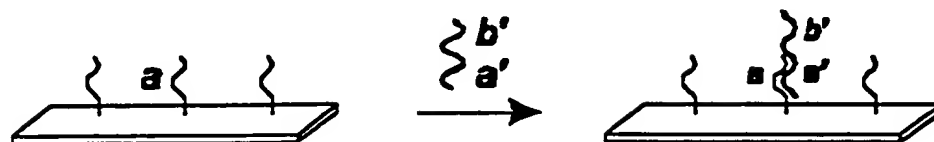
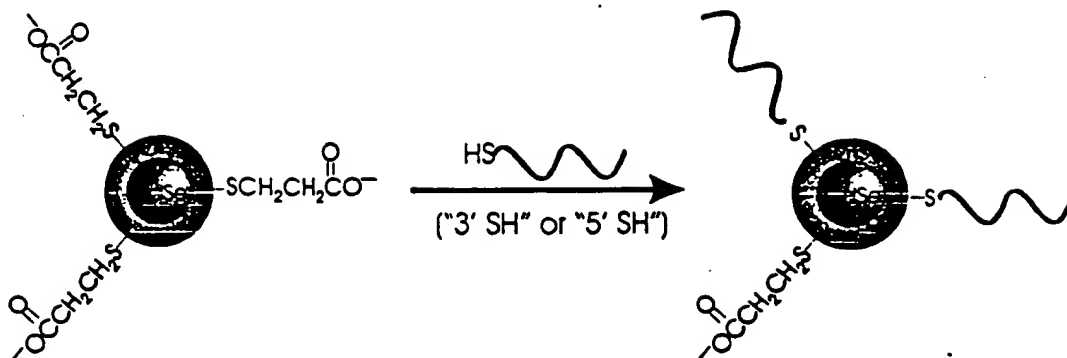


FIGURE 25B

A



B

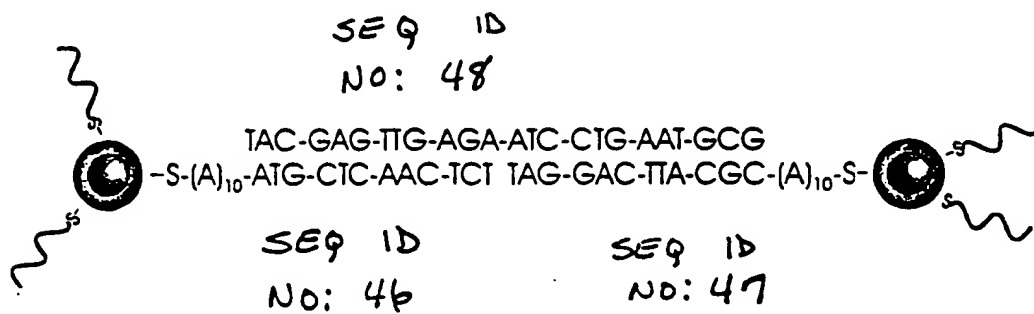


FIGURE 26

TOP SECRET

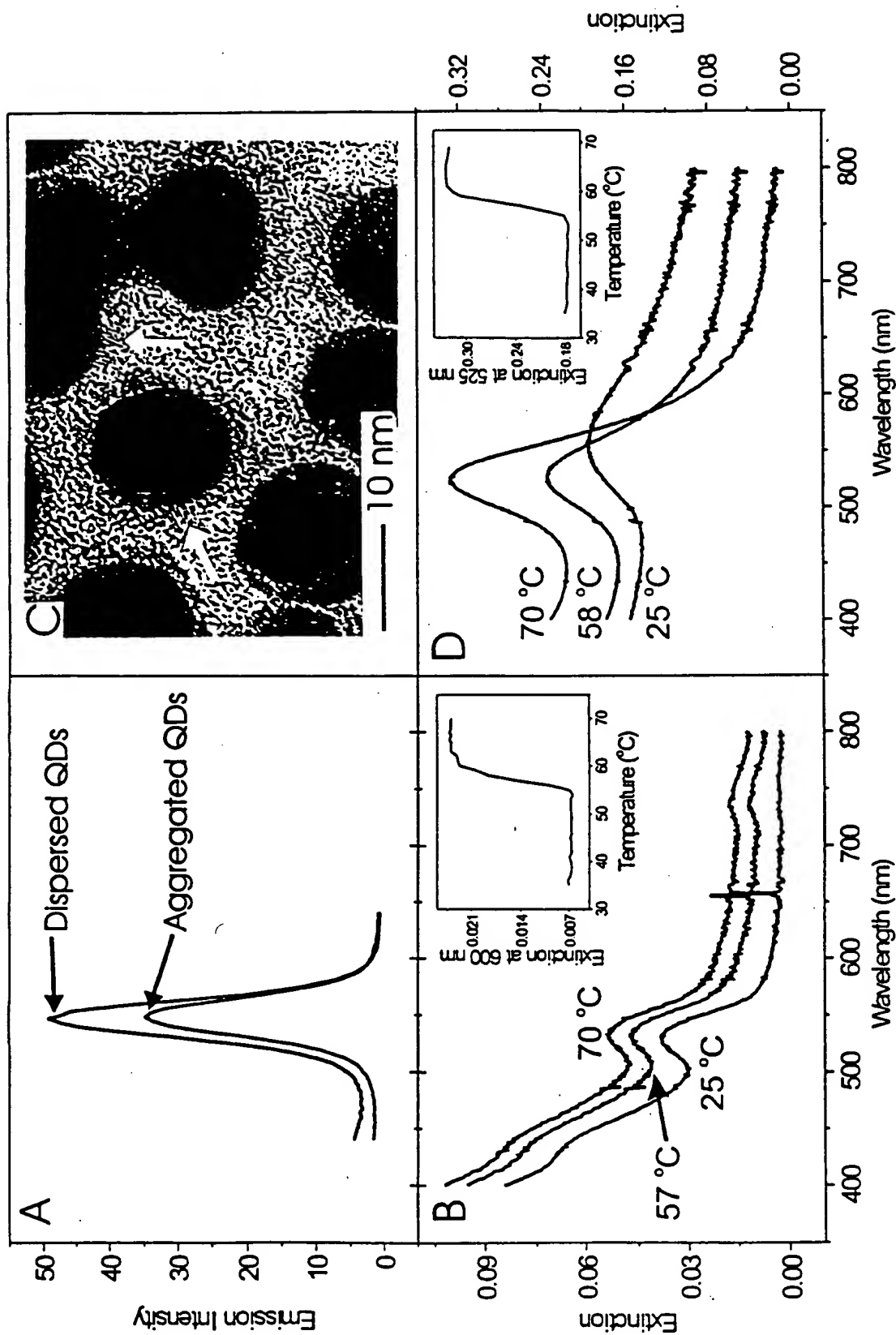


FIGURE 27

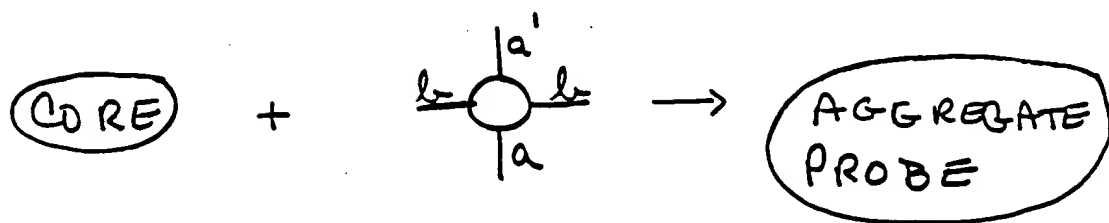
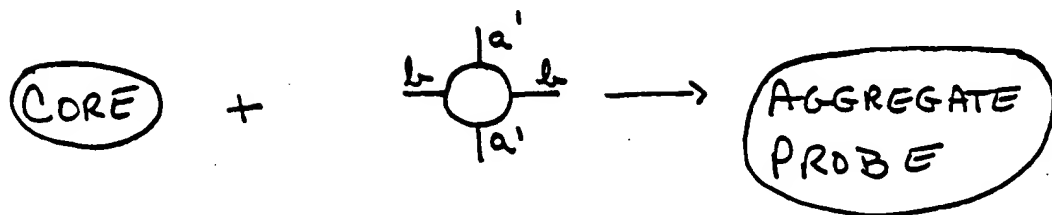
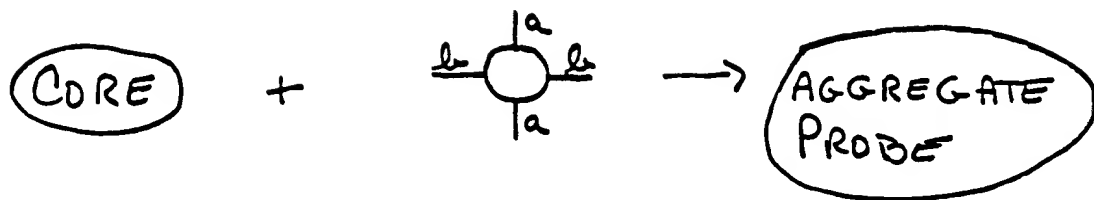
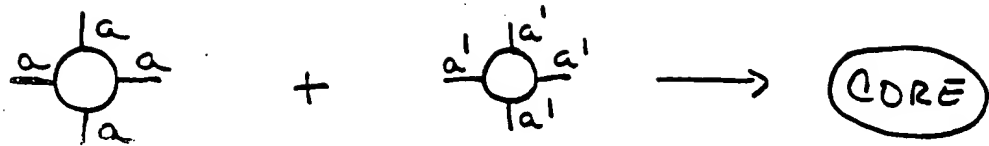
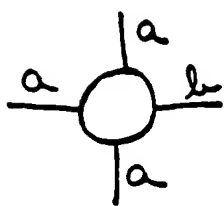
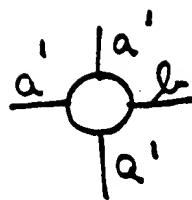


FIGURE 28A



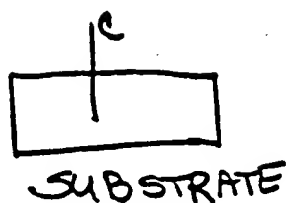
+



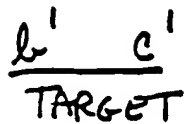
→

AGGREGATE
PROBE

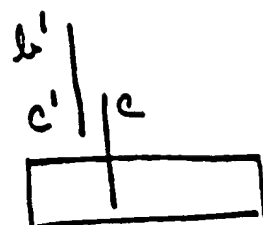
FIGURE 28B



+



→



↓

+ AGGREGATE
PROBE []

↓

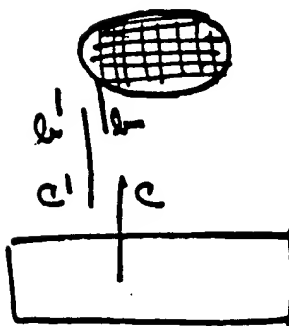
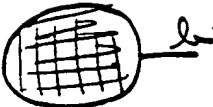


FIG. 28C

TOP SECRET

FOI 4352660


AGGREGATE
PROBE

+ $\frac{b'}{c'}$ TARGET + $\frac{e}{d}$

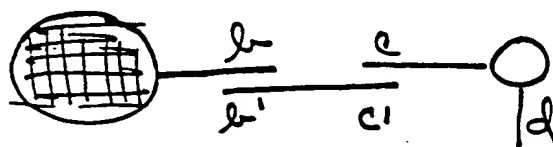
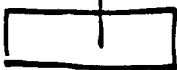
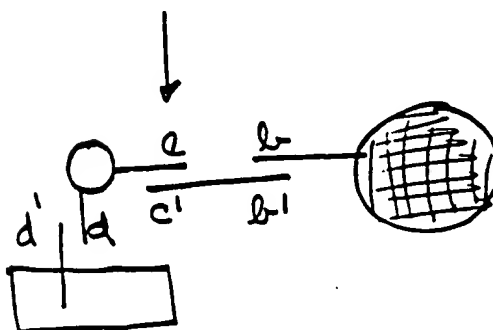


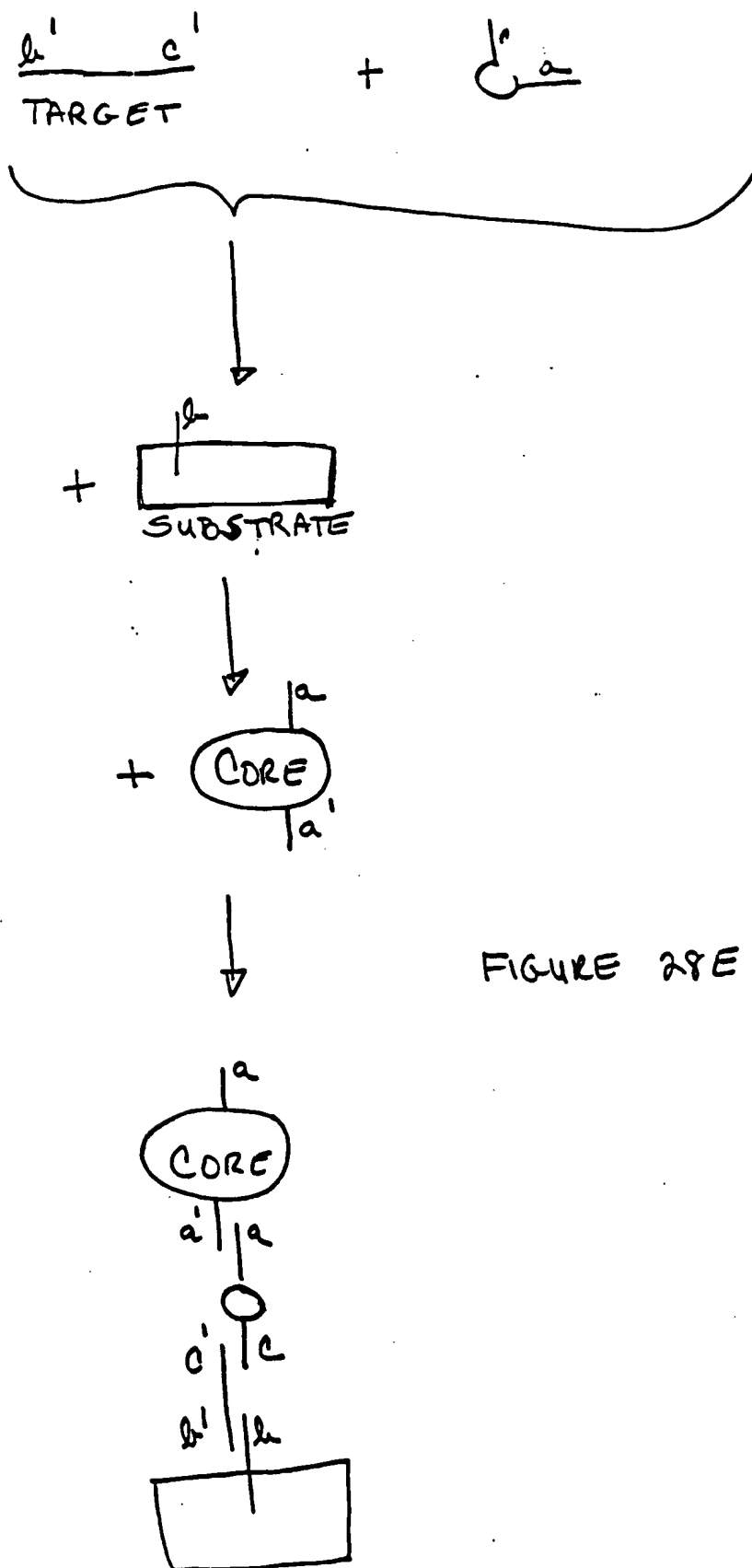
FIG. 29D

REMOVE EXCESS
BY CENTRIFUGATION

+  SUBSTRATE



TTTTT-4BES/660



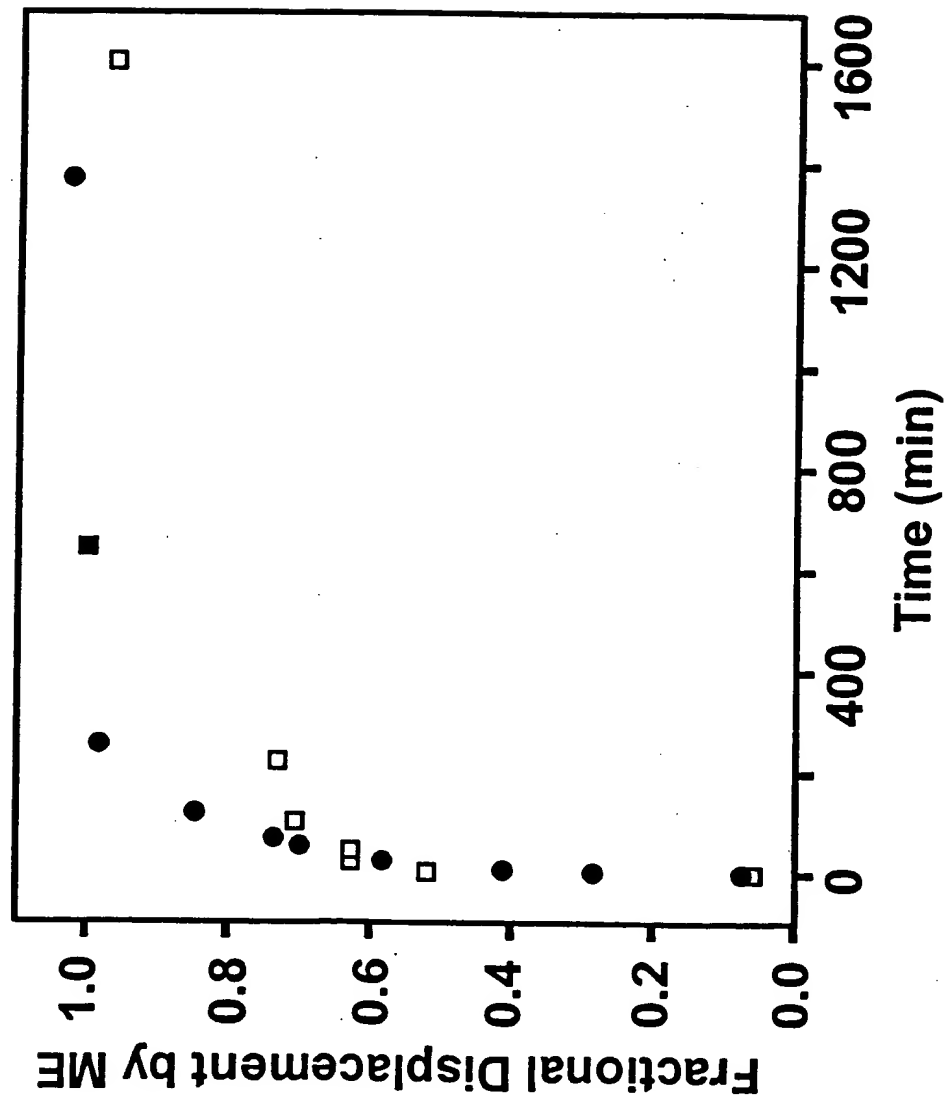


Figure 29

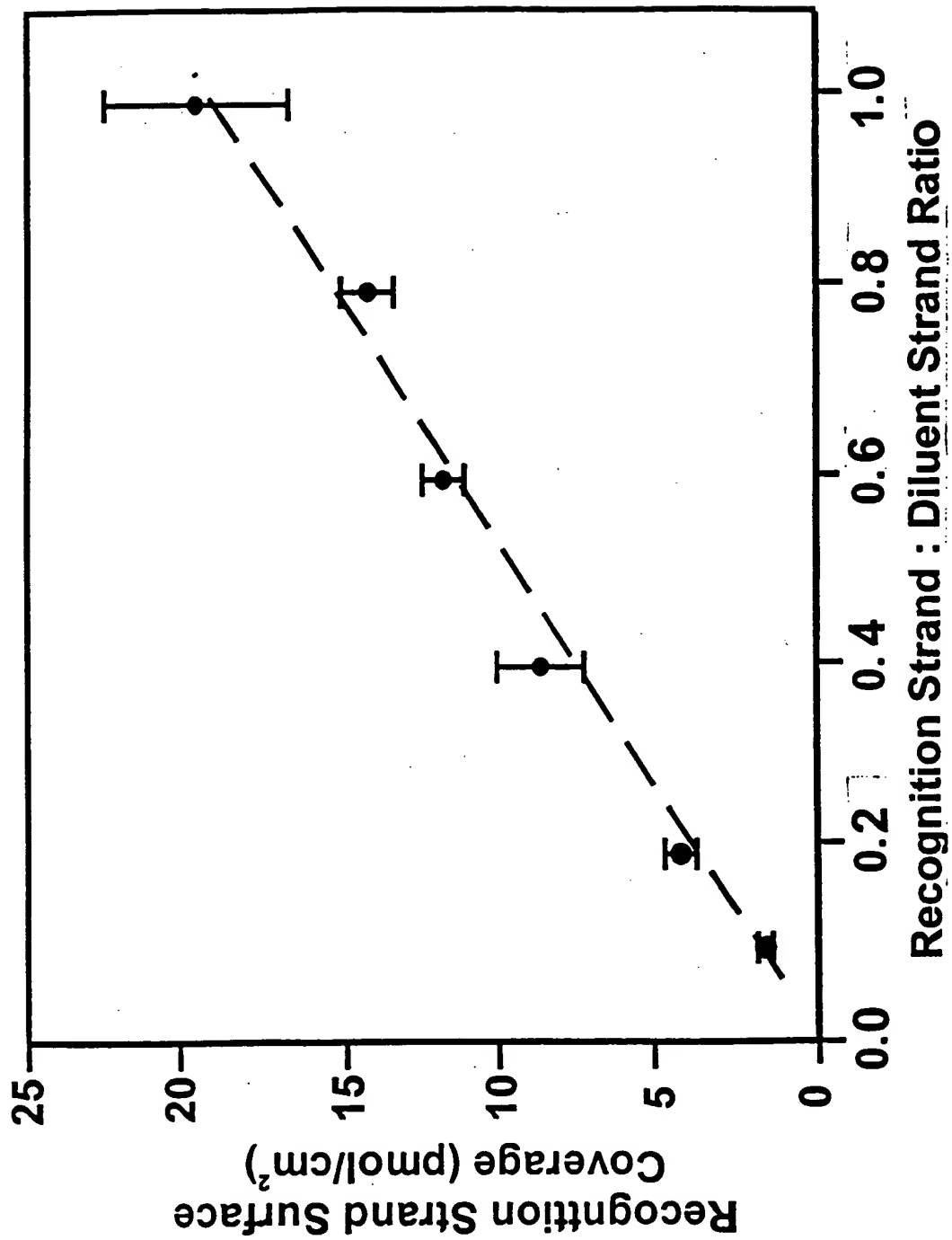


Figure 30

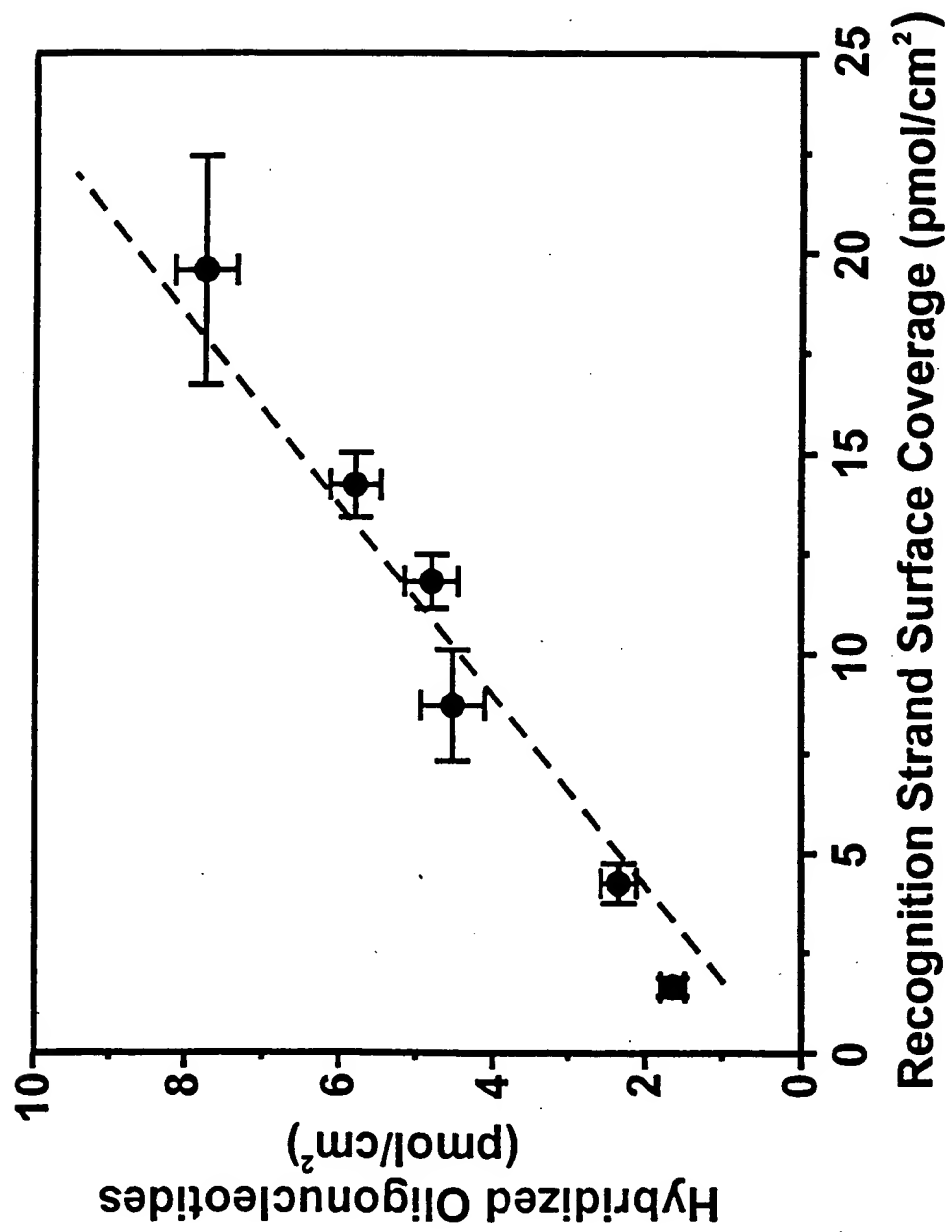
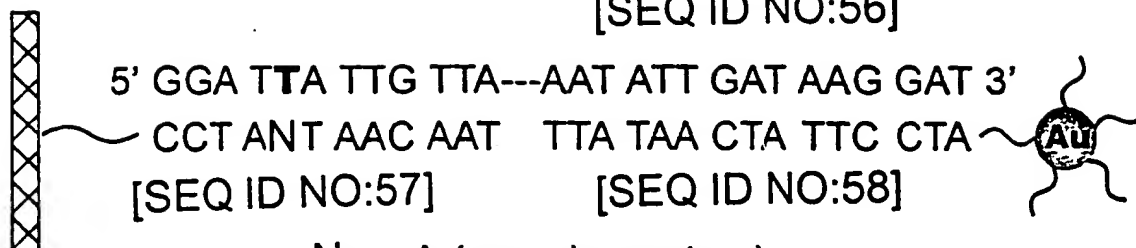


Figure 31

[SEQ ID NO:56]



N = A (complementary),
G,C,T (mismatched)

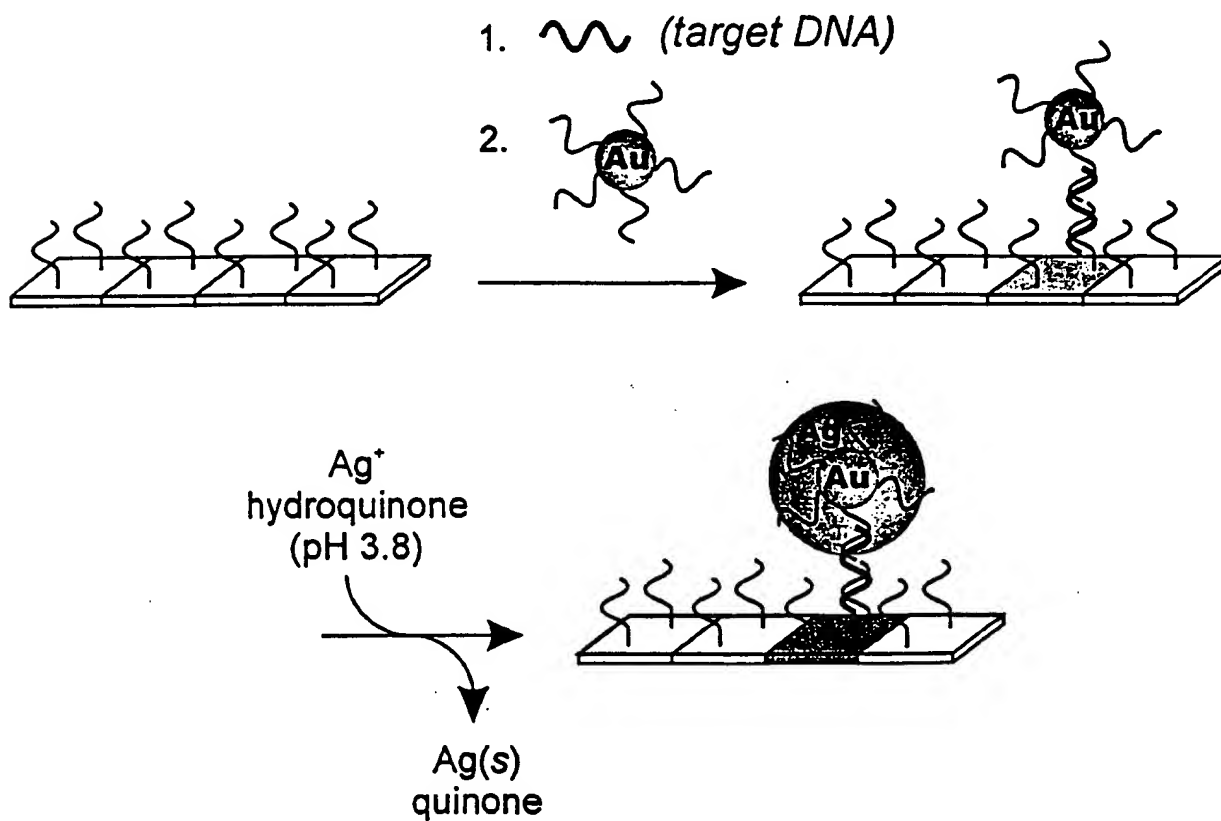


Figure 32

097534-43252660

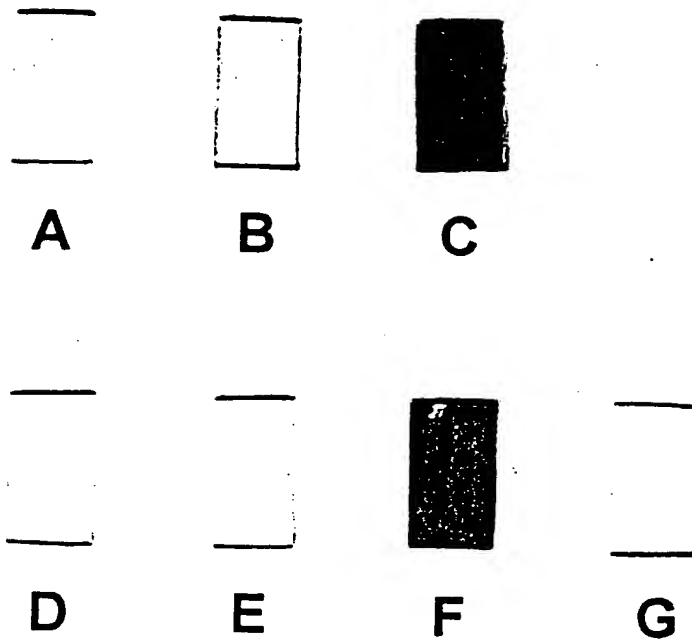


Figure 33

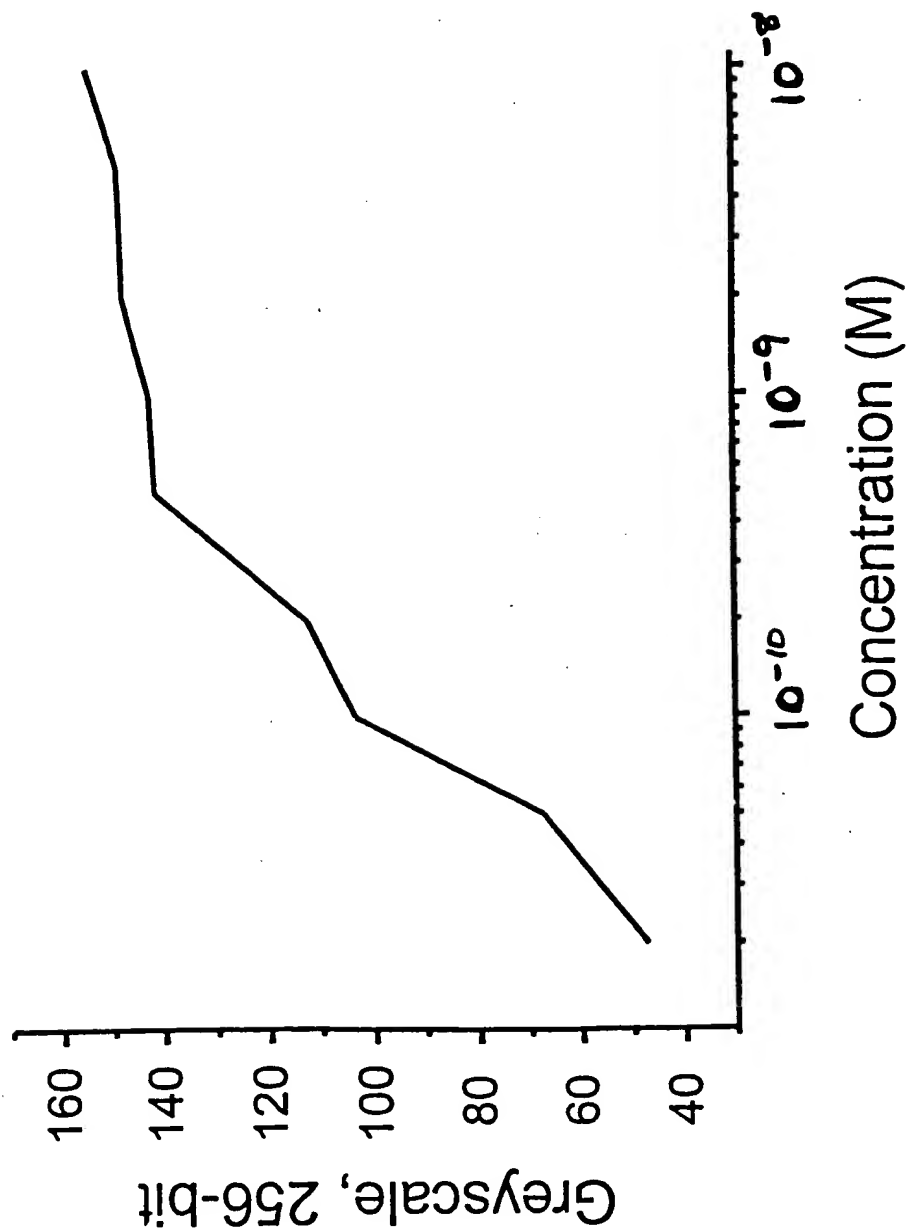


Figure 34

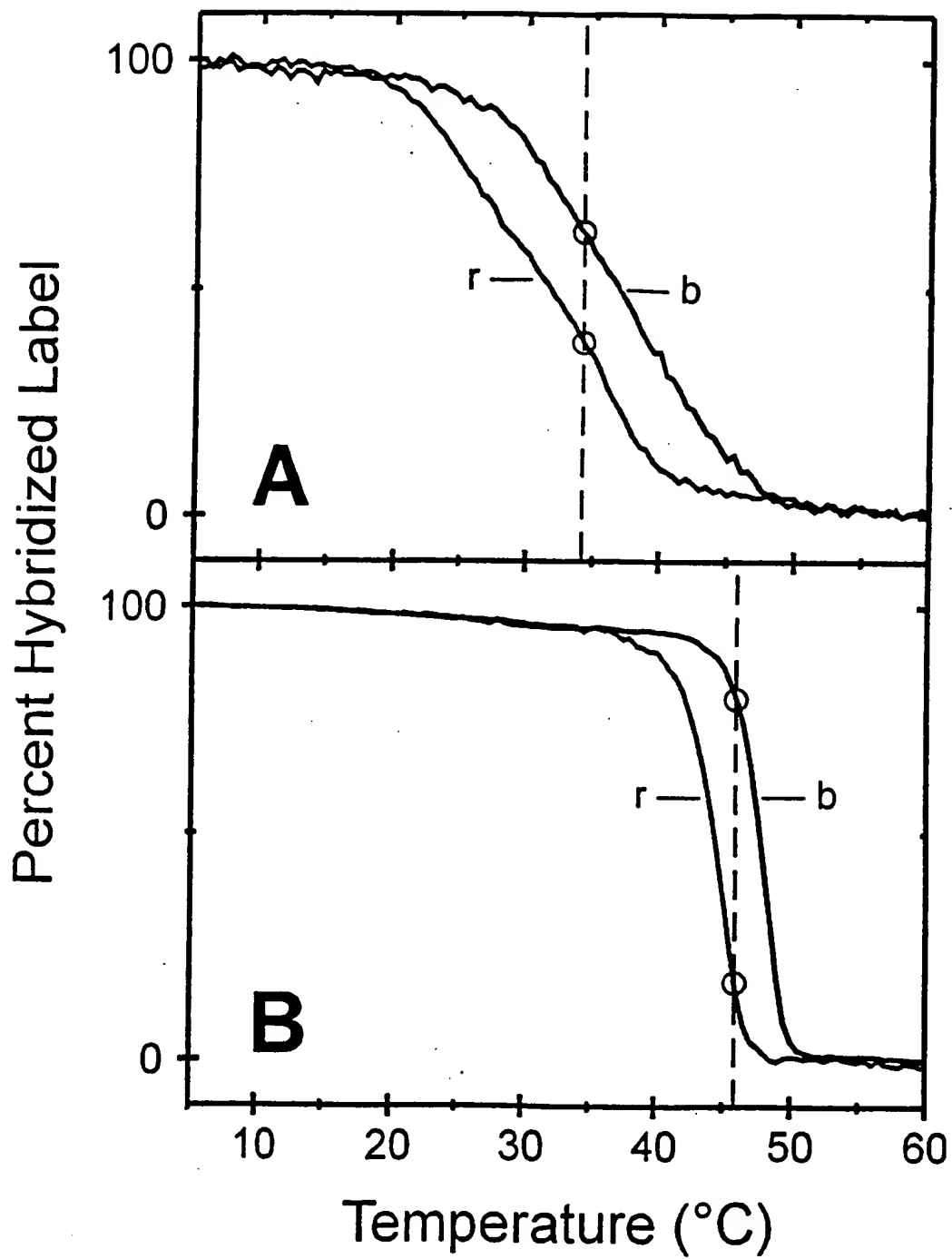


Figure 35

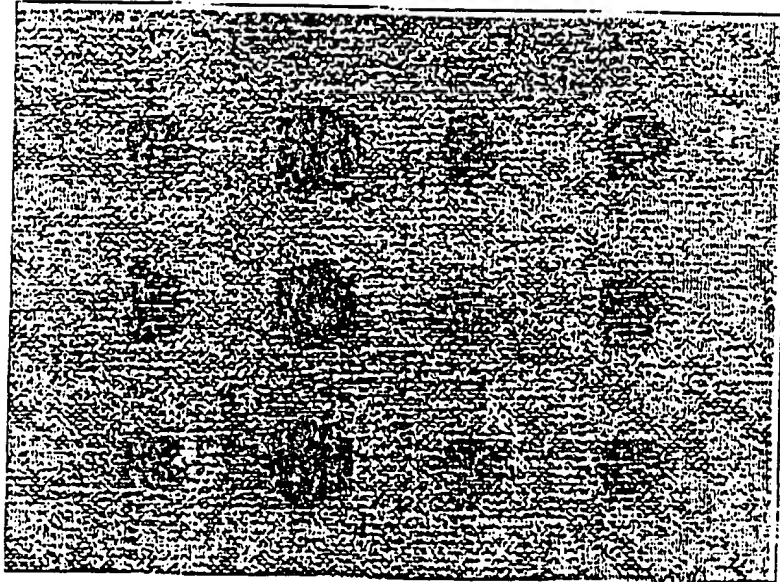
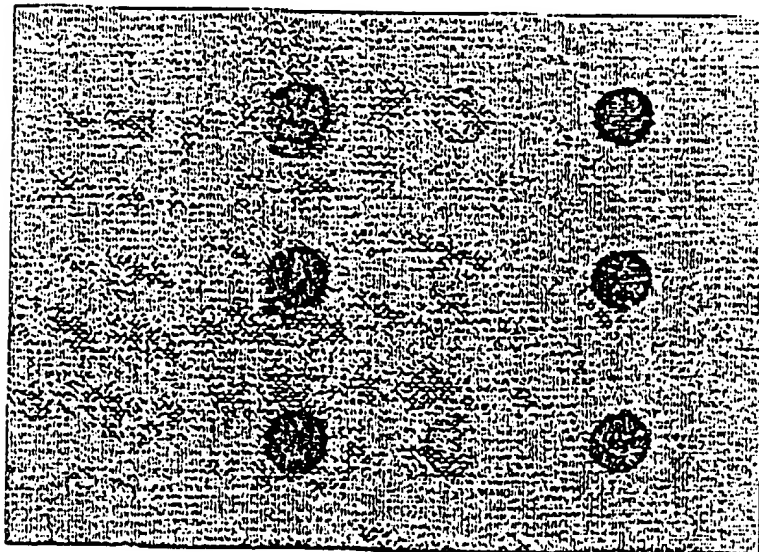


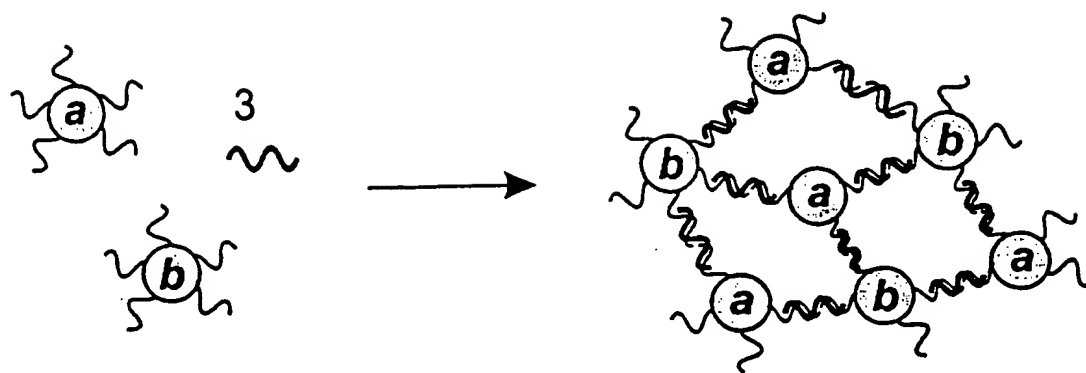
FIG. 36B



C A T G

THE UNIVERSITY OF CHICAGO

A



B

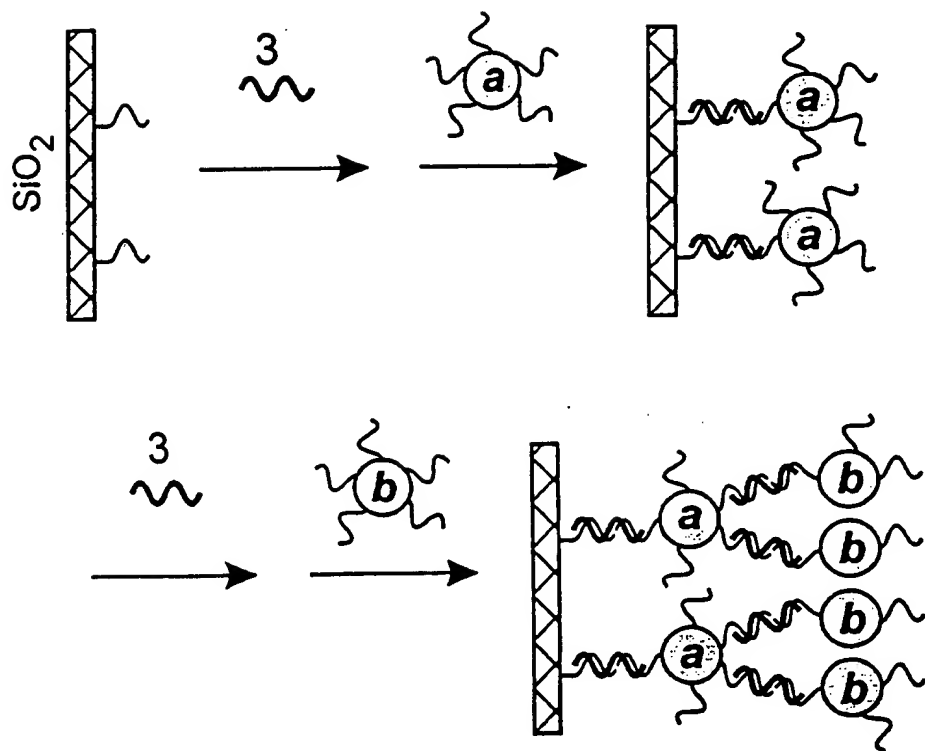


Figure 37

FOOT-13E52660

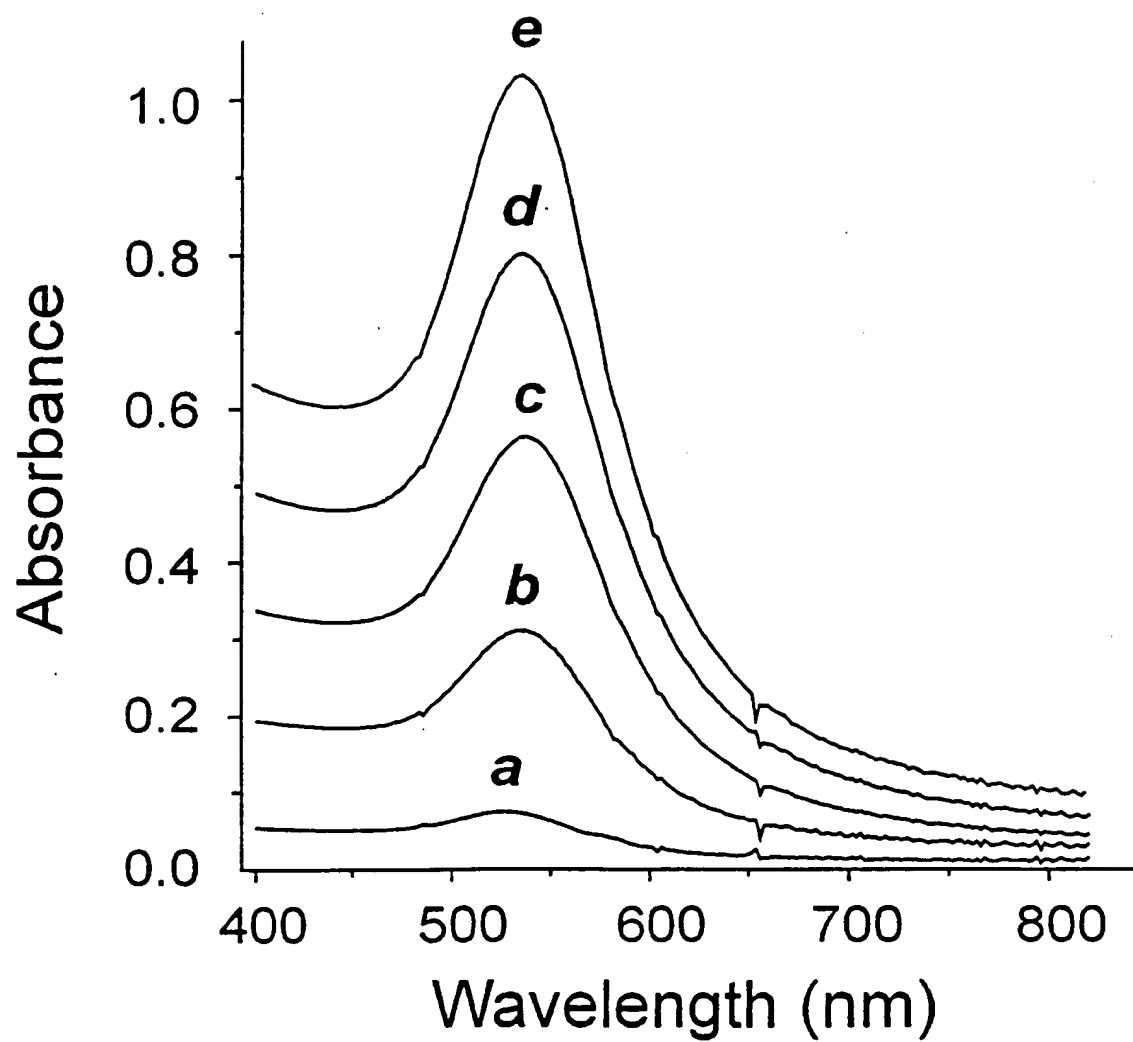


Figure 38A

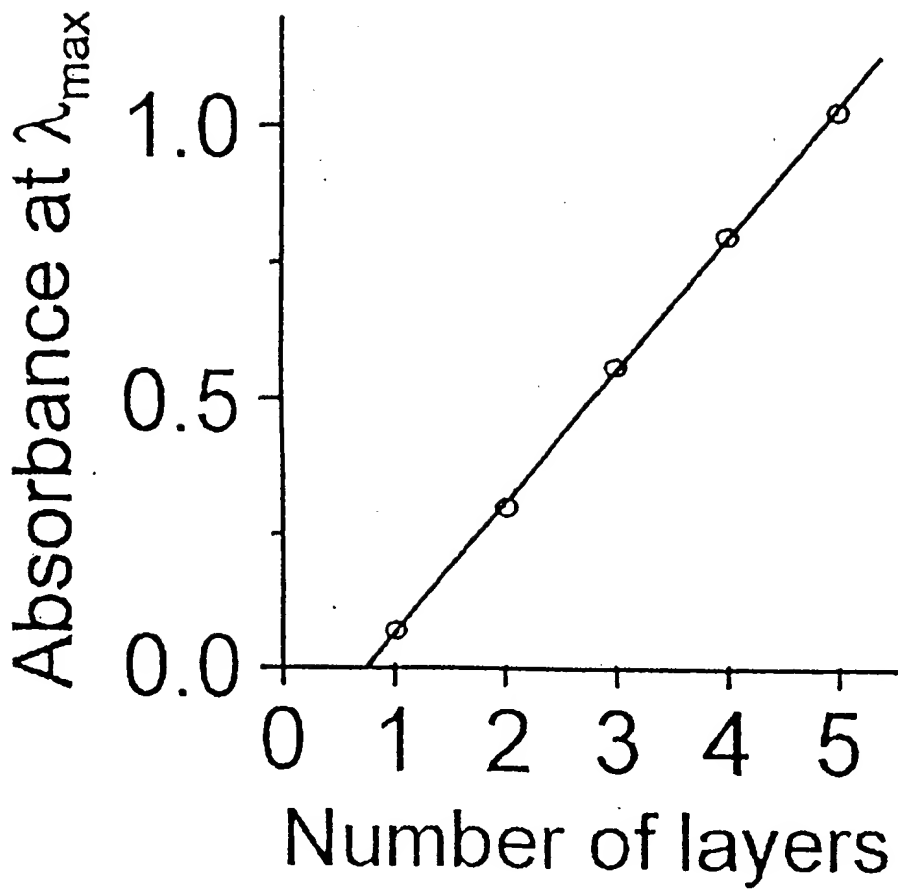


Figure 38B

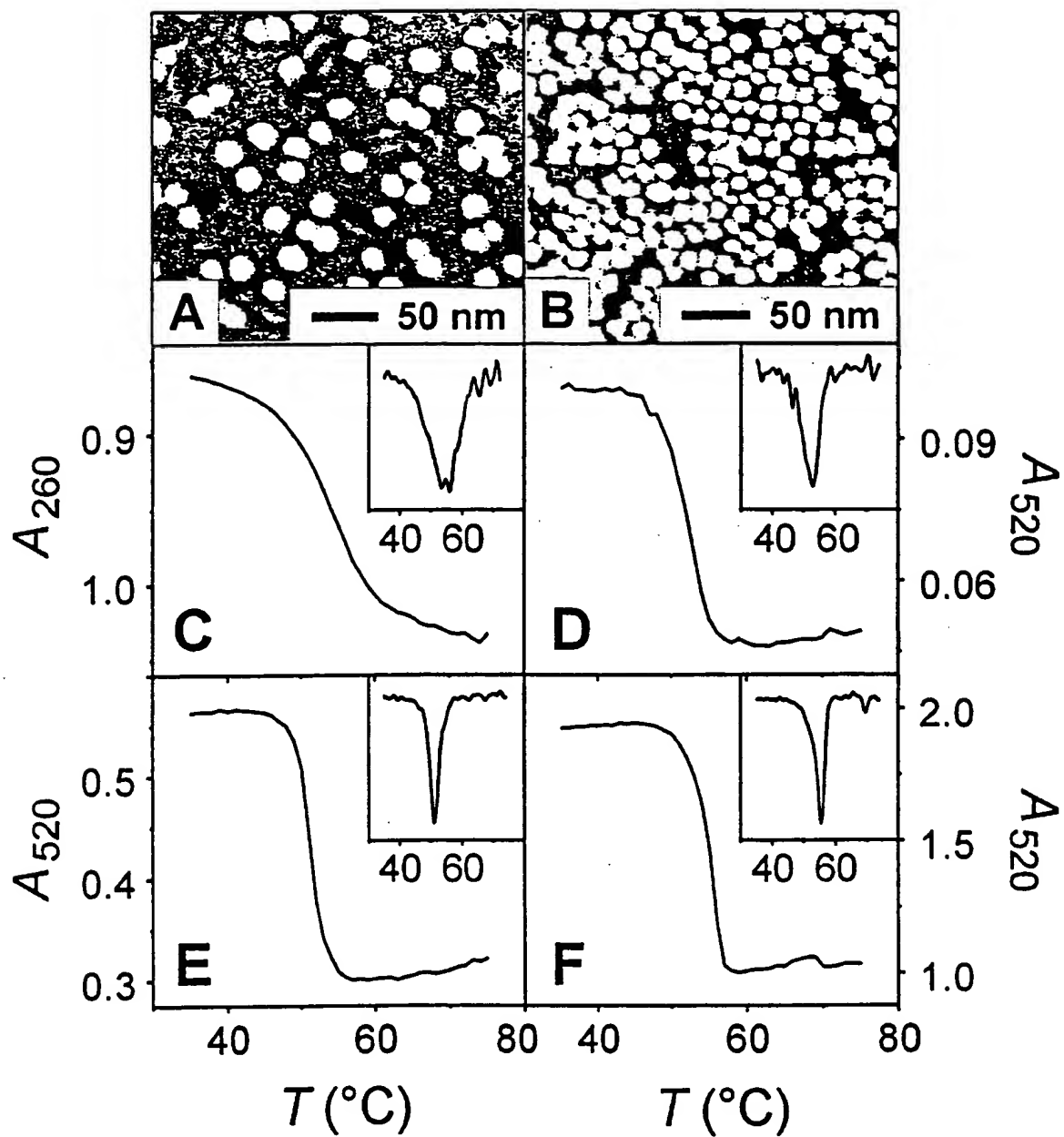


Figure 39

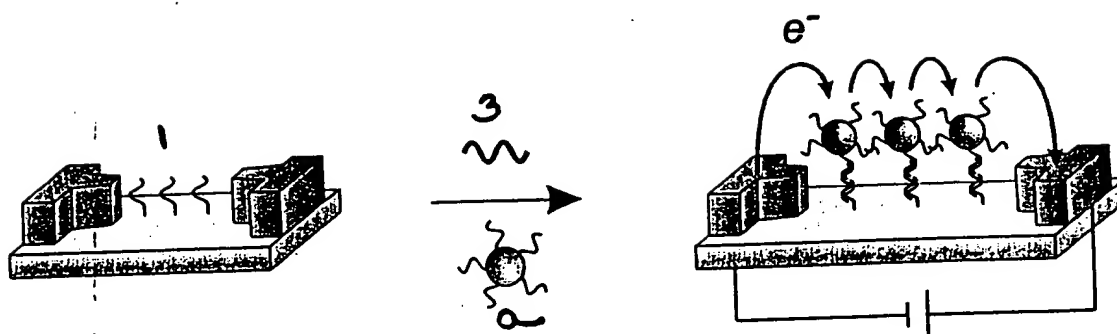
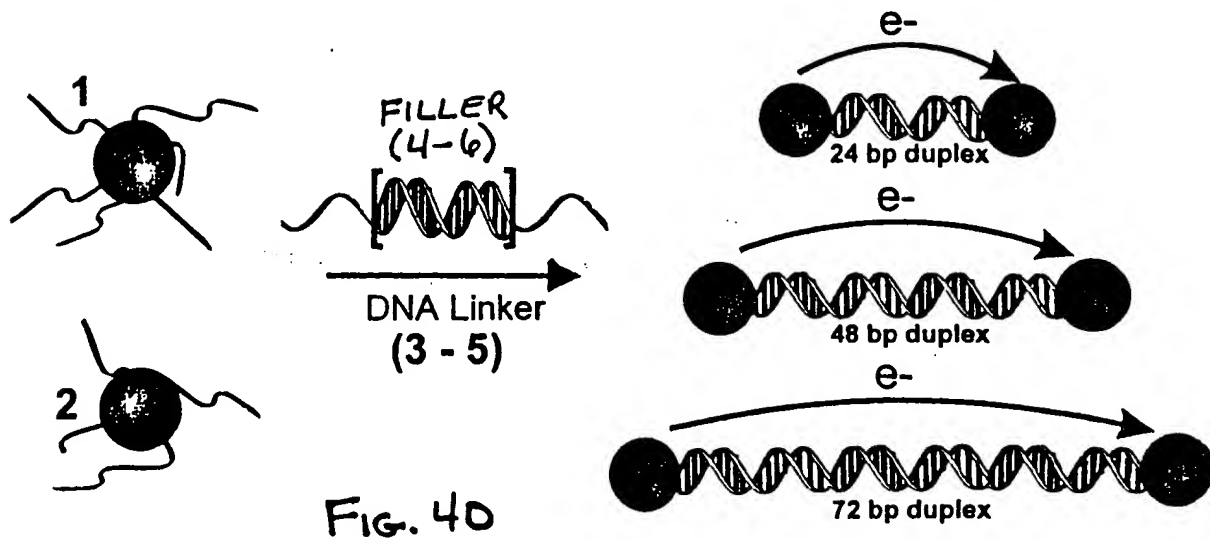


FIG. 41